QUANTIFICATION OF PLASTIC REMNANTS ON TITANIUM SURFACE AFTER INSTRUMENTATION AND EVALUATION OF EFFICACY OF ITS REMOVAL AFTER IRRIGATION BY USING A CONFOCAL MICROSCOPE

Dissertation submitted to

THE TAMILNADU Dr. M.G.R. MEDICAL UNIVERSITY

In partial fulfilment for the Degree of

MASTER OF DENTAL SURGERY

BRANCH II
DEPARTMENT OF PERIODONTICS
MAY 2019

CERTIFICATE

This is to certify that this dissertation titled, **"QUANTIFICATION OF PLASTIC REMNANTS ON TITANIUM SURFACE AFTER INSTRUMENTATION AND EVALUATION OF EFFICACY OF ITS REMOVAL AFTER IRRIGATION BY USING A CONFOCAL MICROSCOPE"** is a bonafide record of work done by **Dr. NIMISHA GOPAL** under our guidance and to our satisfaction, during her postgraduate study period of 2016-2019.

This dissertation is submitted to **THE TAMILNADU Dr. M.G.R. MEDICAL UNIVERSITY** in partial fulfillment for the award of the degree of **MASTER OF DENTAL SURGERY - PERIODONTICS, BRANCH II**. It has not been submitted (partial or full) for the award of any other degree or diploma.

Dr. Koshy Chithresan, MDS,
Professor & Head of the Department,
Department of Periodontics,
Sri Ramakrishna Dental College & Hospital.

Dr. J. Srihari, MDS,
Professor and Guide,
Department of Periodontics,
Sri Ramakrishna Dental College & Hospital.

Dr. V. Prabhakar, MDS,
Principal,
Sri Ramakrishna Dental College & Hospital.

Date:

Place: Coimbatore.

DECLARATION

TITLE OF DISSERTATION	QUANTIFICATION OF PLASTIC REMNANTS ON TITANIUM SURFACE AFTER INSTRUMENTATION AND EVALUATION OF EFFICACY OF ITS REMOVAL AFTER IRRIGATION BY USING A CONFOCAL MICROSCOPE
PLACE OF STUDY	SRI RAMAKRISHNA DENTAL COLLEGE AND HOSPITAL, COIMBATORE-641006.
DURATION OF THE COURSE	3 YEARS
HEAD OF THE DEPARTMENT	Dr. KOSHY CHITHRESAN
NAME OF THE GUIDE	Dr. J. SRIHARI

I hereby declare that no part of the dissertation will be utilized for gaining financial assistance or any promotion without obtaining prior permission of the Principal, Sri Ramakrishna Dental College and Hospital, Coimbatore. In addition, I declare that no part of this work will be published either in print or in electronic media without the permission of the guide who has been actively involved in this dissertation. The author has the right to reserve for publish of work solely with the prior permission of the Principal, Sri Ramakrishna Dental College and Hospital, Coimbatore.

Head of the Department **PG Guide** **Signature of the Candidate**

Principal

Urkund Analysis Result

Analysed Document: THESIS.docx (D46462454)
Submitted: 1/6/2019 6:59:00 PM
Submitted By: nimishakg.123@gmail.com
Significance: 0 %

Sources included in the report:

Instances where selected sources appear:

0

CERTIFICATE – II

This is to certify that this dissertation work titled **"QUANTIFICATION OF PLASTIC REMNANTS ON TITANIUM SURFACE AFTER INSTRUMENTATION AND EVALUATION OF EFFICACY OF ITS REMOVAL AFTER IRRIGATION BY USING A CONFOCAL MICROSCOPE"** of the candidate **Dr. NIMISHA GOPAL** with registration number **241613352** for the award of **MASTER OF DENTAL SURGERY** in the branch of **PERIODONTICS (BRANCH II).** I personally verified the urkund website for the purpose of plagiarism check. I found that the uploaded thesis file contains all pages from introduction to conclusion and the result shows **0%** plagiarism in the dissertation.

Head of the Department **PG Guide**

ACKNOWLEDGEMENTS

Foremost, I thank God Almighty for all the blessings bestowed upon me.

I thank my guide Dr. J.Srihari, MDS, Professor, Department of Periodontics, Sri Ramakrishna Dental College and Hospital, for his able guidance throughout and instilling in me that learning is a continuous and everlasting process. His perfectionist ways have helped me grow in many ways.

I would like to express my whole hearted gratitude to Dr. Koshy Chithresan, MDS, Head of the Department, Department of Periodontics, Sri Ramakrishna Dental College and Hospital for taking out the time and being patient with me throughout my period of dissertation and for helping me understand the subject better.

I thank Dr. Arun Maradi, MDS, Reader, Dr. Praveen Krishna, MDS, Reader, and Dr. Rajesh, MDS, Senior Lecturer, Department of Periodontics, Sri Ramakrishna Dental College and Hospital for the constant support and guidance.

I also thank Dr. Surya, Dr. Vimala and Dr. Cynthia for their support.

I thank Dr. V. Prabhakar, MDS, Principal, Sri Ramakrishna Dental College and Hospital, for providing me with the necessary facilities.

I take this moment to thank my seniors; Dr. Subash, Dr. Fazal Ilahi, Dr. Megha Madhusoodhanan, Dr. K.K.N Saranya and Dr. Asha for their steady support and guidance throughout this period. I thank my juniors; Dr. Sheetal and Dr. M. Indrajit for being supportive throughout. My sincere thanks to Dr. M. Junaid for his expertise in statistics.

Special thanks to Dr. Ritika Chhalani and Ms.Sneha Venkataramana for keeping me sane and for being there always throughout this challenging period.

I express my sincere thanks to Dr. M.R.Ganesh, PhD, Assistant Professor and Ms.Saranya Sekar, MPhil, National Facility for Clinical Trial, Interdisciplinary Institute of Indian System of Medicine (IIISM) at SRM University, Kattankalathur for their time and effort.

I sincerely acknowledge Dr. P.Radhakrishnan, PhD, Professor and Dean of PSG Institute of Advanced Studies and Mr.Sakthi Vignesh, Assistant Professor, Department of Robotics and Automation Engineering, PSG College of Technology for their time and effort.

None of this would have been possible without the support of my family. I thank my parents Mr. K. Gopalan and Mrs.Pushpa Gopal for always standing by me, believing in me and for being my strength. I thank my sister Ms.Nithu Mahendhiran and family for always being my support system in all ways.

Dr. Nimisha Gopal

CONTENTS

LIST OF FIGURES

LIST OF TABLES

LIST OF GRAPHS

GLOSSARY

Abbreviation	**Expansion**
ASTM	American Society for Testing and Materials
AFM	Atomic Force Microscopy
BI	Bleeding Index
BOP	Bleeding On Probing
CA	Citric Acid
CAL	Clinical Attachment Level
CFU	Colony Forming Unit
CHX	Chlorhexidine
CI	Calculus Index
CLSM	Confocal Laser Scanning Microscope
DNA	Deoxyribo Nucleic Acid
EDS	Energy Dispersive X-Ray Spectroscopy
EDTA	Ethylene diamine tetra acetic acid
ERL	Erbium-doped Yttrium Aluminium Garnet Laser
GI	Gingival Index
H_2O_2	Hydrogen Peroxide
HA	Hydroxy apatite
HCL	Hydrochloric acid
HeNe	Helium Neon
ICPMS	Inductively Coupled Plasma Mass Spectrometry
LPS	Lipopolysaccharide

L_r	Waviness
MGI	Modified Gingival Index
MP	Machine Polished
NaCl	Sodium Chloride
PBS	Phosphate Buffered Saline
PD	Probing Depth
PEEK	Polyetheretherketone
PI	Plaque Index
R_a	Average Surface Roughness
RBM	Resorbable Blast Material
RSR	Relative Specular Reflectance
R_z	Maximum Surface Roughness
SEM	Scanning Electron Microscope
SI	Stain Index
SLA	Sand Blasted and acid etched
Ti	Titanium
TIE	Trans mucosal Implant Extensions
TPS	Titanium Plasma Spray
WLC	White Light Confocal profilometry

INTRODUCTION

Complete debridement of plaque and all other decontaminants from the implant surface is of paramount importance for long term success and survival of dental implants. Various techniques have been used which includes, non-surgical and surgical decontamination with mechanical instruments, antimicrobial therapies and/or lasers[1].

Among all these techniques, surface debridement using plastic and titanium curettes appears to be the treatment protocol for peri-implant mucositis and peri-implantitis. It has been proposed that treating the titanium surface with plastic or non-metal tip prevents damage to the implant surface as compared to the use of metal instruments as the use of metal instruments leads to increased roughness of implant surface and increased biofilm formation[2]. But, it has been known that instrumenting with any material softer than titanium may leave remnants on the titanium surface, [3,4] which can influence bacterial attachment, cell attachment and effectiveness of oral hygiene measures, thereby impairing the biocompatibility of the implant surface[5].

To investigate the nature of these effects, it is necessary to ascertain the amount of plastic debris left on various modified titanium surfaces after treatment. Plastic materials have been known to show significant auto fluorescence when visualised under laser irradiation[6]. Based on this background, the current in-vitro study was conducted under non-simulated conditions to quantify the plastic remnants on surface modified titanium after instrumentation with plastic instruments by using a Confocal laser scanning microscope and also investigate the efficacy of removal of these remnants by using air-water spray and 0.2% chlorhexidine irrigation.

AIMS & OBJECTIVES

AIM: The aim of this in-vitro study was to quantify the surface area covered by plastic remnants on different surface modified titanium discs after instrumentation with various plastic instruments and to evaluate the efficacy of the removal of these remnants by using a confocal laser scanning microscope under non-simulated conditions.

OBJECTIVES:

1. To quantify the plastic remnants on titanium discs that are sand blasted, acid etched, SLA treated and polished after instrumentation with various plastic instruments such as PEEK ultrasonic tip (EMS Piezon Systems), carbon composite ultrasonic tip (PH1, Satelec, Suprason) and plastic curette (Columbia 4R/4L, Hu Friedy) by confocal microscopy.
2. To evaluate the efficacy of the removal of these remnants after irrigation with water spray and 0.2% chlorhexidine for 10 seconds by confocal microscopy.
3. To compare the amount of plastic remnants remaining on various surface modified titanium discs after instrumentation and irrigation.

REVIEW OF LITERATURE

INSTRUMENTATION:

An in-vitro study was conducted by Fox et al in 1990 to evaluate the effects of scaling titanium implant surface (IMZ titanium implant system) using titanium alloy curette, a stainless steel curette and a plastic instrument specifically designed for instrumentation of dental implants. Alterations of the surfaces due to instrumentation were evaluated by a Helium Neon (HeNe) laser and was reported as relative specular fluorescence (RSR). It was concluded that plastic instruments produced an insignificant alteration of the titanium implant surface following instrumentation, while metal instruments such as titanium and stainless steel curettes significantly altered the titanium surface[7].

A study was conducted by Dmytryk et al in 1990 to evaluate the effects of scaling titanium implant surfaces with Plastic (Branemark), titanium-alloy (Norton) and stainless steel curette (Hu-Friedy) on fibroblast cell attachment. Counts of attached cells were made at 24 and 72 hours; the implants were then processed for scanning electron microscopy (SEM). It was found that the cell attachment to stainless steel curette instrumented titanium surfaces was significantly reduced compared to untreated control, titanium alloy curette or plastic instrumented surfaces. SEM observations showed that fibroblast on stainless steel instrumented surfaces tended to show a somewhat rounded morphology and a relatively reduced degree of spreading; while fibroblasts on untreated control, plastic, or titanium-alloy instrumented surfaces showed a well-spread, polygonal morphology, which were more typical of fibroblasts in favourable culture conditions. Hence it was concluded that such observations of cell attachment and morphology after instrumentation with plastic and titanium alloy curette are indicative of in-vivo biocompatibility and might have clinical implications for the proper maintenance of titanium dental implants[8].

The cleaning effectiveness of different treatment methods on titanium abutments was evaluated using scanning electron microscope (SEM) by Speelman JA et al in 1992. Titanium abutments were installed on beagle dogs. After 16 weeks of plaque accumulation, the abutments were instrumented using 1) Metal Gracey curettes (LM Dental); 2) Plastic scalers (Nobelpharma); 3) Ultrasonic scalers (Cavitron, Dentsply); 4) Air-polishing (Stainbuster with Prophy-Jet cleaning powder, Dentsply); 5) Rubber cup polishing with pumice and 6) Brushing with a multitufted brush (Butler). It was found that regular rubber cup polishing and regular brushing resulted in highest surface cleanliness while the air polishing showed the lowest cleanliness score. Treatment with metal, plastic and ultrasonic instruments clinically resulted in clean surfaces. The finding that daily brushing resulted in clean surfaces stresses the importance of daily oral hygiene for implant patients. Finally, taking into consideration the cleanliness, the surface roughness and the possible adverse effects on the biocompatibility, it was concluded that plastic scalers may be the instruments of choice for professional debridement of titanium implant surface[9].

In an in-vitro study by Alan Homiak et al in 1992, the surface of titanium implant abutments using light and scanning electron microscope and also the effects of various forms of hygiene prophylaxis instrumentation on the abutments were evaluated. The instruments used were stainless steel scaler (American Dental Mfg,Co) using moderate finger pressure, plastic scaler (Nobelpharma), rubber cup polishing (Densco Prophy Cup) and air powder abrasive unit (Cavi-Jet, Dentsply). The metal scaler system was found to roughen the titanium surface. All other modalities tested appeared to smoothen the titanium surface by removing the surface debris and rounding off the sharp machined grooves present on the untreated abutment surface. These findings suggested that the use of plastic scaler, rubbercup polishing and air powder abrasive system did not harm the titanium surface and could be thus used for debridement[10].

The surface texture of titanium implant abutments after instrumentation with plastic scaler, air-powder abrasive and rubber cup polishing was evaluated in an in-vitro study by Mc Collum et al in 1992. Plaque accumulation was also compared following instrumentation. The untreated control abutments revealed prominent milling marks and slight pits, the plastic scaler was found to slightly smoothen the milling mark and it caused micro scratches. The air-powder abrasive largely obliterated the milling marks and caused surface pitting whereas the rubber cup with polishing removed the milling marks and created a smooth swirl pattern. It was found that all abutments collected plaque and there was no statistically significant difference between the groups. Thus, it was concluded that, for maintenance and prophylaxis, any of these methods may be used without damaging the abutment surface or enhancing plaque accumulation[11].

Brookshire et al in 1993 in an in-vitro study compared the surface quality of both commercially pure titanium and titanium alloy abutments, subjected to various hygiene methods and instruments such as the 1) Implarette scaler - instrument tip was fabricated from a gold palladium alloy with a gold coating; 2) Plasteel scaler (Implacarea, Hu-Friedy) fabricated from a high grade resin called Plasteel; 3) Universal scaler (Steri-Oss Inc.) which was fabricated from graphite fiber; 4) A slow speed handpiece and prophy angle with screw-type rubber cup and tin oxide slurry; 5) The Prophy-Jet air-powder abrasive system (Dentsply). The surfaces were then analysed under a scanning electron microscope (SEM). Results showed that, no significant surface alterations were produced by the air abrasive system. Implarette scaler, Implacare scaler and universal scaler seemed to leave behind some residue or deposits after instrumentation. It was also found that surfaces treated with the Implarette scaler exhibited the most damage among all the hygiene instruments. Hence, from the findings it was concluded that the air abrasive system seemed to produce the least surface alterations as compared to the Implarette scaler, Plasteel scaler and Universal scaler[12].

The effect of modified scaler tips on variously structured titanium surfaces using stereomicroscopy, scanning electron microscopy and laser profilometry was investigated by Ruhling et al in 1994. Instrumentation was done on different implant surfaces such as smooth machined titanium, etched and sandblasted surfaces, titanium-plasma-sprayed (TPS) and hydroxyapatite coated surfaces (HA) using Cavimed-200 ultrasonic scaler with Teflon coated tip, Sonicflex-2000 sonic scaler with Teflon coated tip, Light curette, Implacare curette and metal instruments such as Cavimed-200 ultrasonic scaler with stainless steel tip, Sonicflex-2000 sonic scaler with stainless steel tip, Implarette gold plated curette and stainless steel Gracey curette (Hu-Friedy). The results revealed that no discernible damage was caused by Teflon coated sonic and ultrasonic scalers or implant curettes made of plastic on smooth titanium surfaces. Surface roughness increased with the use of metal instruments on smooth titanium surfaces. Instrument material residues were found on rough implant surfaces. Thus, it was concluded that coating of sonic and ultrasonic scaler tips with Teflon could be used for supragingival and subgingival cleaning of titanium implant surfaces[3].

Kuempel et al in 1995 conducted a study to examine the epithelial growth on titanium surfaces after instrumentation with plastic scaler, stainless steel scaler and gold coated curettes. The discs were then seeded with a microdot of rat gingival cells. At the end of 5 days, the surface area covered by the epithelial cells were then measured. Results showed that gold coated curette exposed surfaces had less epithelial growth compared to stainless steel, plastic and control surfaces. The epithelial surface area coverage did not vary significantly among groups. But, the specific characteristics of the cellular morphology were found to be different among the groups. Thus, it was concluded that the reduced epithelial growth in gold coated curette instrumented discs might be due to the surface contaminants originating from the gold curette[13].

Meschenmoser et al in 1996 assessed quantitatively and qualitatively effects of various instruments such as stainless steel curette (Schweickhardt); plastic curette (Nobelpharma); a prototype of pure titanium curette, an air abrasive polishing system (Airflow II, EMS) and an ultrasonic system (Cavitron) on titanium abutments. The surface structures were compared with scanning electron microscope (SEM), profilometry and Confocal laser scanning microscope. Evaluation revealed surface alterations for all instruments and systems except the plastic curette which did not roughen the surface. The steel curette and the ultrasonic system proved to be totally unsuitable for cleaning titanium abutments. Even though plastic curette did not roughen the surface, the effectiveness of plastic curette for removing hard calculus and the resultant plastic debris on the implant surface were not evaluated in this study[14].

Surface alterations on titanium implant necks following different prophylaxis procedures such as ultrasonic scaler, Plastic tip ultrasonic scaler, Stainless steel curette, Titanium curette, Teflon curette, Air powered system, Abrasive rubber cups, polishing rubber cup and brush was evaluated by Matarasso et al in 1996. SEM and laser prophylometer analysis was done to measure the roughness in terms of average surface roughness (R_a) and maximum surface roughness (R_z). Results showed that the use of ultrasonic scaler, stainless steel curette, titanium curette and air jet polishing increased the implant surface roughness as compared to controls whereas abrasive rubber cups increased the implant surface smoothness. Use of rubber cup polishing, brush polishing, Teflon curette, plastic curette and plastic tip scaler left the implant surface unaltered[15].

Hallmon et al in 1996 compared the effects of metallic, non-metallic and sonic instrumentation on titanium abutment surface in-vitro, using scanning electron microscopic (SEM) examination. The instruments used were Stainless steel Gracey curette (Hu-Friedy), Implacare plastic curette (Hu-Friedy), Plastic curette (Steri-Oss), Plastic curette (Implant Support), Sonic scaler with metal tip (Titan – S) and Sonic scaler with plastic tip (Dynatip). The highest surface alteration was seen with the Implarette scaler followed by sonic scaler, Gracey curette, Dynatip and Steri-Oss. The Implacare and Implant Support non-metallic scaler had the least surface alteration. It was concluded that the Implacare and Implant support non-metallic (plastic) scalers appear to be the instruments of choice for debridement of titanium abutment surfaces if preservation of surface integrity is the primary objective[16].

Mengel et al in 1998 examined the work traces left by various instruments such as Titanium curette (Deppeler SA), Gracey curette (Hu-Friedy), Plastic curette (Nobel Biocare), Rubbercup with Zircate prophypaste (Dentsply), Cavitron Jet ultrasonic scaler with universal insert (Dentsply), Cavitron Jet air polishing nozzle with Prophy-Jet cleaning powder (Dentsply), Densonic sonic scaler with SofTip disposable prophy tip (Dentsply) and Densonic sonic scaler with universal tip (Dentsply) on implants and abutments by scanning electron microscope and determined the quantity of substance removal by optical laser profilometry. It was found that the Gracey curette, the Cavitron Jet ultrasonic scaler and the Densonic scaler with universal tip left moderate to pronounced work traces and caused increased substance removal followed by the titanium curette and the Densonic sonic scaler with SofTip disposable prophy tip which left slight working traces The rubber cup, the plastic curette and the Cavitron Jet air polishing system caused no visible change to the implant surfaces and caused the least substance removal and thus can be suitable for cleaning implant surfaces[17].

Augthun et al in 1998 examined the effect of specific cleaning procedures such as Plastic curette (DIA 238), Metal curette (Hu-Friedy), Diamond polishing device (Perioset/blue), Ultrasonic scaler (Satelec), Air powder spray with sodium hydrocarbonate solution (Plaque Sweep) and 0.1% CHX solution rinse on the surface of 3 implant types with different coatings and shapes (plasma sprayed, hydroxyapatite coated implants and smooth titanium surface screws) using SEM. The air powder abrasive system, CHX rinse and curettage with the plastic instrument caused little or no surface damage in all but hydroxyapatite coated fixtures. The growth of vital cells on contaminated implants was also observed after treatment. It was found that implants sprayed with the air- abrasive system had the most vital cells. Hence it was concluded that, the use of plastic scalers and air abrasive system had the least damaging effect on plasma coated and smooth titanium implant surfaces[18].

The effects of Er: YAG laser (ERL) and the Vector ultrasonic system on the biocompatibility of titanium implants with four different surfaces (sand-blasted and acid-etched (SLA), titanium plasma-sprayed (TPS), machine-polished (MP) and hydroxyapatite-coated (HA)) in cultures of human osteoblast-like cells was investigated by Schwarz et al in 2002. Cells were counted using a reflected light microscope and the cell density per mm^2 was calculated. Additionally, cell morphology and surface alterations of the titanium discs after treatment were investigated using SEM. It was found that the highest number of cells per mm^2 were seen on SLA surfaces, followed by the TPS and MP surfaces. The HA- coated surfaces showed the least cell density per mm^2. In the laser-treated groups, no thermal side effects such as melting or loss of porosity were observed. However, all surfaces treated with the Vector system showed conspicuous surface damage and deposits of used carbon fibres. Hence it was concluded that Er:YAG laser did not damage titanium surfaces and subsequently did not influence the attachment rate of SAOS-2 cells[4].

Sato et al in 2004 compared the effects of a new ultrasonic scaler with carbon tip (Vector), a conventional ultrasonic scaler with plastic tip (Satelec) and a plastic scaler on titanium surfaces. The roughness was measured with a Profilometer and observed by SEM. It was found that the rate of debris removal by the Vector scaler and the conventional ultrasonic scaler were higher than the plastic scaler. There were no significant differences in surface roughness among the 3 instruments. Hence, it was concluded that the new ultrasonic scaler and conventional ultrasonic scaler were useful for removing artificial debris and produced no significant damage to titanium surfaces compared to plastic scalers[19].

Karring et al in 2004 compared the effectiveness of treatment of peri-implantitis with a Vector system and carbon composite curette. Instrumentation was done at baseline and at the end of 3 months. Plaque, BOP and PPD were recorded on all implant surfaces at baseline, and after 3 and 6 months. At the end of 6 months, it was found that four of the Vector treated sites and one site treated with carbon curettes had stopped to bleed. Thus, it was concluded that there was greater reduction in the number of sites with BOP following treatment with the Vector system than following instrumentation with carbon fiber curettes, but the difference was not found to be statistically significant[20].

In an in-vitro study by Ramaglia et al in 2006, the effects of different instrumentations used in the treatment of peri-implantitis on implant surfaces coated with hydroxyapatite or titanium plasma spray (TPS) was investigated. The implant surfaces were treated with a stainless steel Gracey curette (Premier), plastic curette

(Implant scaler, Premier), ultrasonic scaler tip (Satelec) and air-powder-water spray (Airflow). Profilometry and SEM were used to examine the instrumented surfaces. It was found that the plastic curette and air-powder-water spray induced less implant surface alterations, though these instrumentations left deposits on the surface that may affect, in-vivo, the tissue healing process[21].

Kawashima et al in 2007 evaluated the treatment of titanium implants with ultrasonic scalers with Carbon tip (Vector), Plastic tip (Satelec) or Metallic tip (Enac). The abutment surface characteristics were examined after instrumentation using SEM and laser profilometer. The amounts of remaining plaque and calculus were estimated using the modified remaining plaque and calculus score developed by Speelman. The surface alterations were evaluated using the modified roughness score developed by Hallmon. The abutments treated with the Vector scaler and plastic scaler had essentially clean and smooth surfaces. No calculus was observed, although some small particles of amorphous material were seen. The abutments treated with the metallic tip scaler had irregularities and defects but had clean surfaces with no calculus. Thus it was concluded that piezoelectric scalers with non-metal tips were suitable for use in dental implant maintenance[22].

Mann et al in 2011 conducted a study to assess the effect of plastic covered ultrasonic scalers on titanium implant surface. The inserts used included a TFI 10 metallic tip and a plastic coated ultrasonic implant insert (SofTip, Dentsply) driven by a Cavitron SPS 30 kHz ultrasound generator. The plastic cover of the modified insert probe was screwed into place on an adapted metallic scaler. The implant surfaces were then scanned using laser profilometer and SEM. It was found that the metal scalers produced defects in titanium implant surfaces whereas plastic coated probes caused minimal damage to implant surface. It was also found that the plastic coated scalers had a polishing action and left plastic deposits behind on the implant surface[23].

A study to evaluate the safety and efficiency of novel ultrasonic scaler tips, conventional steel tips and plastic tips on titanium surface. Mechanical instrumentation was carried out using conventional scalers with a novel metallic implant tip (Cetatech), a plastic headed tip (EMS), Plastic tip (Satelec) and a conventional stainless steel tip (EMS) was conducted by Baek et al in 2011. The instrumented surface samples were viewed with a SEM and surface profile was investigated using an atomic force microscope (AFM). SEM images on surfaces scaled by the novel metallic implant tip and the EMS plastic tip showed no marked differences in surface morphology. Surfaces instrumented using the conventional stainless steel tip showed higher surface roughness[24].

Sahm et al in 2011 conducted a study to evaluate the effectiveness of air abrasive device and carbon curette with antiseptic therapy and CHX for non-surgical treatment of peri-implantitis. At the end of 6 months it was found that both the treatment procedures resulted in comparable but limited CAL gains and air abrasive device was found to be associated with significantly higher BOP reductions than carbon curette with antiseptic therapy and CHX[25].

Schmage et al in 2012 evaluated the effects of variety of implant cleaning instruments on different implant surfaces, especially surface roughness and cleaning efficiency. Biofilm layers of *Streptococcus mutans* were cultivated on titanium discs with four different surface modifications (polished, grit blasted, acid etched, and acid etched/grit blasted). The instruments used were 1)Plastic curette (Hu-Friedy), 2)Carbon curette (Hawe Neos), 3)Prophylaxis brush (Sonic Flex brush), 4)Rubber cup (Hawe

Neos), 5)Sonic driven PEEK plastic tip (Sonic- Flex Clean), 6)Ultrasonic driven PEEK tip (Piezon Master), 7)Ultrasonic driven carbon composite tip (Satelec), 8)Vector system, 9)Air polishing (ProphyJet) and 10) Er:YAG laser. Results showed that the surface roughness for the acid etched surfaces, polished and the grit blasted surfaces showed no significant differences between the different cleaning instruments compared to control groups. Significantly lower surface roughness was seen on grit blasted/acid etched implant surfaces following use of prophylaxis brush and plastic curette, followed by sonic driven PEEK tip, Vector system, ultrasonic driven PEEK tip, rubber cup, Er:YAG, air polishing and carbon curette[26].

Park et al in 2012 evaluated the effects of oral hygiene instruments including various types of ultrasonic tips such as 1) ultrasonic scaler with metal tip (EMS Piezon Systems), 2) ultrasonic scaler with plastic tip (EMS Piezon Systems), 3) ultrasonic scaler with metal tip (Suprason; Satelec), 4) ultrasonic scaler with plastic tip (PH1; Satelec), and 5) brush (Implant care brush) (Implant Care; TePe) in simulated clinical settings and brushing with dentifrice on machined and SLA titanium surface with confocal microscopy. It was concluded that metal or plastic ultrasonic scaler tips may be applied as usual to treat the SLA surface without increasing the irregularities on the titanium surfaces. However, in case of machined surfaces, ultrasonic metal tips cannot be recommended because the surface becomes rougher after treatment[27].

Fakhravar et al in 2012 investigated the surface roughness on the apical collar of implant abutments caused by probing and scaling instruments. The instruments used were 1) UNC -15 metal probe, 2) Periowise plastic probe, 3) Mc Call SM 17/18 metal scaler and 4) Universal plastic scaler (Hu-Friedy). Surface roughness was assessed with a contact profilometer. The plastic probe and plastic scaler did not significantly affect the abutment surface, but left behind residues. This debris may be firmly attached to the surface of the abutment both through mechanical attachment to the machining grooves on the abutment and through electrostatic forces based on charge differences between the plastic particles and the metal surface. This debris then creates large "positive" artifacts on the surface, thus contributing significantly to surface roughness. On the other hand, the metal probe seems to have had limited or no effects on the abutment surface. Thus, it was concluded that probing around implant abutments with a metal probe seemed to have no effect on the surface but, instrumentation with scalers (plastic and metal) and plastic probe may cause surface roughness[28].

Unursaikhan et al in 2012 characterized changes in the roughness of titanium surfaces treated by various scaling instruments such as piezoelectric ultrasonic scaler with a newly developed metallic tip (B & L Biotech), a piezoelectric ultrasonic scaler with a conventional tip (EMS), a piezoelectric root planer ultrasonic scaler with a conventional tip (EMS), and a plastic hand curette (Hu-Friedy). The treated titanium surfaces were observed by SEM and a profilometer. Most of the procedures increased Rz, the exception was treatment with the plastic hand curette. Hence it was concluded that, the roughness values (Ra and Rz) of the titanium surfaces increased in all, except plastic hand curette and the newly developed metallic tip groups, which showed decreased roughness relative to the untreated control group[29].

Park et al in 2013 compared the effects of different instruments on surface roughness and removal of bacteria from Resorbable blast material (RBM) titanium implant discs. The instruments used were 1) ultrasonic scaler with metal tip (EMS), 2) ultrasonic scaler with plastic tip (EMS), 3)Ultrasonic scaler with metal tip (Satelec), 4) ultrasonic tip with carbon tip (Satelec) and 5) Toothbrush (Implant care). The changes in surface roughness were measured using confocal microscopy. A statistically significant decrease in arithmetic mean value of RBM surfaces (R_a) was observed after treatment with an ultrasonic scaler with a metal tip. The discs were incubated with bacteria and instruments were used to remove the bacteria. The amount of remaining bacteria was evaluated using a crystal violet assay. It was found that the metal tip and brushing was more efficient in removing bacteria from the contaminated titanium surface according to the crystal violet assay[30].

Park et al in 2013 conducted a study to evaluate the removal of *Porphyromonas gingivalis* from SLA titanium discs after the discs were instrumented by various ultrasonic scaler tips such as 1) ultrasonic scaler with metal tip (EMS), 2) ultrasonic scaler with plastic tip (EMS), 3)Ultrasonic scaler with metal tip (Satelec) 4) ultrasonic tip with carbon tip (Satelec) 5) Toothbrush (Implant care) using crystal violet assay and SEM and also to assess the change in surface roughness after the treated discs. The smoothest surfaces were produced by EMS metal curette tip and toothbrush followed by EMS plastic tip, Satelec plastic tip and Satelec metal tip. Quantification of remaining bacteria was also assessed. Lowest number of adhering bacteria was noted with metal tip groups. Highest adherence of bacteria was seen in the brushing group even though brushing with dentifrice seemed to produce the surface with lowest roughness[31].

Blasi et al in 2014 conducted a study to compare the efficacy of different instruments on biofilm removal from implant supported restorations. Patients with peri-implant mucositis was treated with ultrasonic scaler with plastic tip, titanium curette, airflow with glycine powder and rubber cup with polishing paste. Results showed that there was no significant difference between the four groups in inflammatory status reduction of peri-implant mucosa. Thus it was concluded that non-surgical therapy was effective in reducing peri-implant mucositis. Although a higher efficacy was seen with ultrasonic scaler with plastic tip and rubber cup with polishing paste when compared to titanium curettes or airflow with glycine powder[32].

Schmage et al in 2014 evaluated the effects of implant prophylaxis instruments on polished and acid etched implant surfaces. Biofilm layers of *Streptococcus mutans* were grown on the titanium discs. They were instrumented using Plastic curette (Hu-Friedy), Carbon curette (Hawe Neos), Prophylaxis brush (Sonic-Flex Clean -KaVo), Rubber cup (Hawe Cleanic, Hawe Neos), Sonic driven PEEK plastic tip (Sonic- Flex clean – Kavo), Ultrasonic driven PEEK plastic tip (Piezon Master 400 with Pi-instrument EMS) and Air polishing (Dentsply). After cleaning, the surfaces with remaining bacteria were assessed by light microscopy. The best cleaning effectiveness with less than 4% residual biofilm was observed with sonic and ultrasonic oscillating PEEK tips and air polishing followed by prophylaxis brush and rubber cup. The worst cleaning effectiveness was obtained with the manual plastic and carbon curette, with up to 18 % residual biofilm[33].

Smith et al in 2015 evaluated in-vitro topographical and composition changes after instrumentation using ultrasonic scaler with metal tip and plastic coated PEEK tip (EMS) on machined and moderately roughened titanium surfaces. Surface topography analysis was performed using SEM and confocal laser scanning microscopy (CLSM). Surface element composition and rinsing solutions were evaluated using energy-dispersive spectroscopy (EDS) and trace elemental analysis using inductively coupled plasma mass spectrometry (ICPMS). Results demonstrated severe surface topographical alterations with metallic tips and mild to moderate changes for plastic tip instrumented sites. ICPMS analysis of rinsing solutions identified titanium and other metal traces with the use of metallic tips and mainly titanium and carbon when plastic tips were used. Thus, it was concluded that the use of metallic tips produces more pronounced changes than the plastic tips[34].

Bertoldi et al in 2015 evaluated changes to titanium implants smooth surfaces after instrumentation using low vacuum scanning electron microscope (LV-SEM) and white light confocal (WLC) profilometry. The surfaces were instrumented using 1) Stainless steel Gracey curette (Hu-Friedy), 2) Titanium Langer curette, 3) ultrasonic device with probe covered with plastic tip (Cavitron Softip). It was found that the surfaces were significantly roughened after use of stainless steel curette compared to titanium curette and plastic tip. Moreover, an accumulation of titanium after treatment with stainless steel curette and plastic debris, after plastic tip ultrasonic device treatment, inside the implant-abutment gap was recorded. Thus, it was concluded that careful use of titanium curette produced only a slight smooth surface alteration even over prolonged treatments, without debris production that could endanger implant preservation unlike the plastic curette[35].

Ronay et al in 2015 assessed the cleaning potential of commonly used implant debridement methods, stimulating non-surgical peri-implantitis therapy in-vitro. Ink stained implants were instrumented using 1) A Gracey steel curette (Hu-Friedy), 2) An ultrasonic device with a steel tip (PiezoLED Scaler Tip 201, KaVo), 3) An air powder abrasive device (AIRFLOW Master, EMS) with glycine powder and a nozzle for subgingival use. Micro-morphologic surface changes were analysed using SEM. SEM evaluation displayed considerable surface alterations after instrumentation with Gracey curettes and ultrasonic devices, whereas glycine powder did not result in any surface alterations. Among all the treatments, the air powder abrasive device showed a superior cleaning potential[36].

Al-Hashedi et al in 2016 evaluated the effect of four commonly used decontamination methods such as 1) Metal curettes (Hu-Friedy), 2) plastic curettes (Implacare, Hu-Friedy), 3) titanium brush 4) Er:YAG laser on the surface chemistry and bacterial load of biofilm-contained Ti implants. Evaluation was done using SEM and X-ray photoelectron spectroscopy. The presence and viability of bacteria were evaluated with live-dead assays. The organic layer tightly adhered to Ti surfaces could not be completely removed with any of the methods assessed. Ti brushes achieved greater elimination of organic contaminants and bacteria than curettes and Er:YAG laser; however, none of them were able to restore the original surface chemistry. Thus it was concluded that Ti brushes were more effective than curettes (metal or plastic) and Er:YAG laser in decontaminating Ti implant surfaces and Er:YAG laser was more effective than curettes and Ti brushes in killing the biofilm bacteria[37].

Schmidt et al in 2016 evaluated surface characteristics of implants after using different instruments and biofilm formation following instrumentation under SEM. The implants were instrumented using 1) stainless steel curette (Hu-Friedy) 2) titanium curettes; air-polisher using glycine-based 3) perio (PP) or 4) soft (SP) powders or 5) erythritol powder (EP); and an ultrasonic device using 6) stainless steel (PS) or 7) plastic-coated instruments (PI). Implants were then rinsed and subjected twice to bacterial colonisation with *Streptococcus gordonii* (2 hours) and a mixed culture (*S. gordonii, Actinomyces naeslundii, Fusobacterium nucleatum, Porphyromonas gingivalis and Tannerella forsythia*; 24 hours). Quantitative scoring of the photographs revealed that Stainless steel curette caused a significantly rougher surface followed by air polishing with Perio powder, soft powder and erythritol powder, titanium curette, ultrasonic device with metal tip and the least was with ultrasonic device with plastic tip. No significant differences in the surface characteristics (except for stainless steel curette) or bacterial colonization based on one-time instrumentation was concluded[38].

IRRIGANTS USED ON IMPLANT SURFACES:

Zablotsky et al in 1992 conducted a study to determine the nature of residual hydroxyapatite (HA) coated implant surface after treatment with various chemotherapeutic agents such as citric acid, CHX, hydrogen peroxide, tetracycline HCl, stannous fluoride, polymyxin B and a prototype plastic Cavitron tip. Implant surfaces after treatment were evaluated SEM and spectrometrically using Energy dispersive spectrometry (EDS) and X ray diffraction. All treatments left either microscopic residues or loss of surface roughness when viewed on SEM. Results suggested that both citric acid and the plastic cavitron tip had the least residual lipopolysaccharide (LPS) counts. On the other hand, CHX and stannous fluoride left significantly greater amounts of LPS on surfaces than controls. Thus, it was concluded that treating the infected HA-coated implant surface with a 30- 60 seconds application of citric acid was more beneficial in detoxifying the HA coating prior to regenerative procedures as compared to CHX[39].

Dennison et al in 1994 in an in-vitro study investigated the relationship between implant surfaces and decontamination treatments to determine which treatment was the most effective for treating a particular implant surface. The implants used in the study were press fit cylindrical titanium units with machined, plasma sprayed and hydroxyapatite-coated surfaces. Implants were coated with 125I-LPS and treated by burnishing with a cotton pellet soaked in water, citric acid solution (CA), or 0.12% CHX; or treated with an air-powder abrasive (AIR). It was found that the air abrasives were equal to or better than the other treatments on all implant surfaces treated. Air abrasive treatment was the most effective of the four treatments on plasma-sprayed implants, was equally as effective as citric acid on hydroxyapatite-coated implants, and was equally as effective as water or CHX on machined implants. CHX was found to have a poor ability to remove the endotoxin from the hydroxyapatite surface. This may be related to the soponifying effect of the detergents found within CHX. Thus, CHX was found to better distribute the endotoxin on the implant surface, rather than removing the endotoxin from the surface. Thus it was concluded that CHX tended to function poorly when used to detoxify contaminated implant surfaces[40].

Felo et al in 1997 conducted a study to evaluate the effect of irrigation with 0.06% CHX using a powered oral irrigator (Water Pik) with a special subgingival irrigating tip (Pik Pocket Subgingival Tip) compared to rinsing with 0.12% CHX once daily in peri-implant maintenance. Modified Gingival Index (MGI), Plaque Index (PI), Bleeding Index (BI), Calculus Index (CI) and Stain Index (SI) was measured at 3 months. Intergroup comparisons showed that CHX irrigation produced statistically significantly greater reduction than CHX rinsing in the PI, MGI, and SI. The irrigation group also showed a greater reduction in BI and CI than the rinsing group but these differences were not statistically significant[41].

Porras et al in 2002 conducted a study to determine the clinical effects of CHX on peri-implant mucositis at 1 and 3 months as determined by the MPI, mSBI, CAL and PD. The effect of CHX on the microbial flora of mucositic lesions was also evaluated using DNA probes. Test group included mechanical cleansing with rubber cups and polishing paste, plastic scalers for removing calculus and oral hygiene instructions, supplemented by local irrigation with 0.12% CHX using a plastic syringe and the topical application of CHX gel. Control group received only mechanical cleansing and oral hygiene instructions. It was concluded that both modalities of treatment were effective in reducing peri-implant mucositis and probing depths and improving attachment levels[42].

Trejo et al in 2006 performed an experiment to evaluate clinically and histologically the effect of mechanical therapy with or without antiseptic therapy on peri-implant mucositis lesions in nine cynomolgus monkeys. Peri-implant lesions were induced by placing silk ligatures and allowing plaque to accumulate for 6 weeks. The monkeys were randomly assigned to three treatment groups: group A, mechanical cleansing only using rubber cups and polishing paste; group B, mechanical cleansing and local irrigation with 0.12% CHX and application of 0.2% CHX gel; and group C, control, no treatment. It was concluded that for pockets of 3-4mm, (1) mechanical therapy alone or combined with CHX resulted in the clinical resolution of peri-implant mucositis lesions, (2) histologically, both treatments resulted in minimal inflammation compatible with health, and (3) the mechanical effect alone was sufficient to achieve clinical and histologic resolution of mucositis lesions[43].

Sennhenn- Kirchner et al in 2009 conducted a study to evaluate the efficacy of four common antimicrobial agents in the reduction of aerobic bacteria grown in biofilms on rough titanium samples. The solutions investigated contained CHX, essential oil, octenidine, or citric acid. Results showed significant differences in antimicrobial efficacy for the different regimens depending on bacterial species or even the subtype as compared to untreated controls. The reduction rates achieved varied from 30% after 2 minutes of rinsing with CHX to 99.8% after 8 minutes of rinsing with octenidine. Thus it was concluded that the irrigation regimens reduced bacterial colonization in a mature biofilm grown intraorally on rough titanium surfaces. The highest absolute reduction was achieved after 8 minutes, but only the 2-minute reduction rates are significant for clinical practice. Taking this into consideration, the distinct decontamination efficacy of octenidine and citric acid was found to be evident[44].

Gosau et al in 2009 conducted a human in-vivo study to evaluate the efficacy of six antimicrobial agents on the surface decontamination of an oral biofilm attached to titanium implants. The specimens were treated with six antimicrobial agents such as 1) sodium hypochlorite 2) 3% hydrogen peroxide 3) 0.2 % CHX 4) Plax 5) Listerine and 6) 40% citric acid for 1 minute. After which the total bacterial load was quantified and analysed with fluorescence microscopy. Results suggested a significantly lower ratio between dead and total adhering bacteria (bactericidal effect) after incubation with control phosphate-buffered saline (PBS), Plax mouth rinse and citric acid than after incubation in sodium hypochlorite, hydrogen peroxide, CHX and Listerine[45].

Muhling et al in 2010 conducted a study to investigate whether an additional full mouth disinfection would result in a greater clinical and microbiological improvement compared to sole mechanical debridement within one session in patients with peri-implant mucositis and treated chronic periodontitis. After randomized assignment to a test and a control group, patients received a one-stage full-mouth scaling with or without CHX. Clinical and microbiological examination was performed at baseline, after 1, 2, 4 and 8 months. Additional microbial samples were taken 24 h after treatment. Microbiological analysis was performed by real-time PCR. Results showed that both treatment modalities led to an improvement of the clinical parameters and a temporary reduction of the microflora at implants with mucositis, but without significant inter-group differences after 8 months[46].

Ntrouka et al in 2010 conducted a study to assess the effectiveness of different chemotherapeutic agents on biofilm-contaminated titanium surfaces. In experiment 1, *Streptococcus mutans* biofilms grown on titanium discs were treated with (1) EDTA, (2) citric acid (CA), (3) cetylpyridium chloride, (4) Ardox-X, (5) H_2O_2, (6) CHX and (7) sterile water. The three most potent chemotherapeutic agents were selected, and their effectiveness in killing polymicrobial biofilms grown on titanium discs was tested in experiment 2. The biofilms were treated for 5 minutes either with one of the monotherapies, (1) CA, (2) Ardox-X or (3) H_2O_2, or with combined therapies of (4) Ardox-X (2.5min), followed by CA (2.5min) or (5) H_2O_2 (2.5min), followed by CA (2.5min). Results showed that H_2O_2, Ardox-X and CA killed significantly more *S. mutans* compared to the other treatments. H_2O_2 and CA removed significantly more protein than water. CA and the combination treatments were significantly more effective against the polymicrobial biofilms than CHX, H_2O_2 and Ardox-X. Thus it was concluded that among the chemicals tested, CA demonstrated the greatest decontamination capacity with respect to both the killing and the removal of biofilm cells[47].

Burgers et al in 2012 conducted a study to evaluate the antibacterial efficacy of six different topical antiseptics on three test microorganisms attached to titanium implant specimens. Machined pure titanium specimens were used in the study. The titanium discs were incubated either in *Candida albicans, Streptococcus sanguinis*, or *Staphylococcus epidermidis* for 2 hours. The specimens were then treated with different topical antiseptics for 60 s (1% sodium hypochlorite, 3% H_2O_2, 0.2% CHX, 40% citric acid, Plax, or Listerine) and with sterile saline as control. Remaining vital fungi were quantified by means of a bioluminometric assay and the bacterial load and the viability of adhering *S. epidermidis* and *S. sanguinis* by live or dead cell labelling in combination with fluorescence microscopy. It was found that sodium hypochlorite was effective against all three species, whereas hydrogen peroxide was solely effective against *C. albicans*. CHX and Listerine showed antimicrobial activity against *S. sanguinis* and *C. albicans* and citric acid and Plax against both tested bacteria[48].

Charalampakis et al in 2014 conducted a study to investigate the combined effect of mechanical and chemical cleansing on a 4 day biofilm grown intra orally on titanium discs with different surface characteristics. Four titanium discs with four different surface characteristics (OsseoSpeed^{TM}, TiOblast^{TM}, experimental and turned surface) were used. After 4 days of biofilm growth, titanium discs from the right side of the splint were cleaned for 5 seconds each, using three strokes with a cotton pellet soaked in saline while the discs from the left side were cleaned in the same manner but using cotton pellets soaked in CHX. The titanium discs were then processed for SEM analysis. It was found that the combination of mechanical and chemical cleansing was ineffective in complete biofilm removal from all four titanium discs. It was found that Listerine had the largest effect against anaerobes and smallest effect on aerobes (streptococci). Whereas, CHX had better antimicrobial efficacy on streptococci aerobes[49].

Yang et al in 2015 conducted a study to quantify the surface area covered by plastic remnants after instrumentation with various plastic instruments and also to evaluate the efficacy of removal of these remnants after irrigation. The discs were instrumented with 1) Plastic curette (Hu-Friedy), 2) Carbon tip (Satelec) and 3) PEEK (Polyetherether ketone tip (EMS)). The discs were then cleaned with 0.2% CHX soaked cotton pellets and air water spray for 10 seconds. It was found that 10-20% of the surface was covered with plastic remnants irrespective of the instrument used. These remnants could not be completely removed with the air water spray or CHX soaked pellet. Thus, it was concluded that plastic remnants remained after instrumentation, regardless of the irrigation used[5].

Lee et al in 2018 conducted a study to investigate the factors that interfere with osteoblast adhesion to contaminated titanium surfaces after different surface treatments.

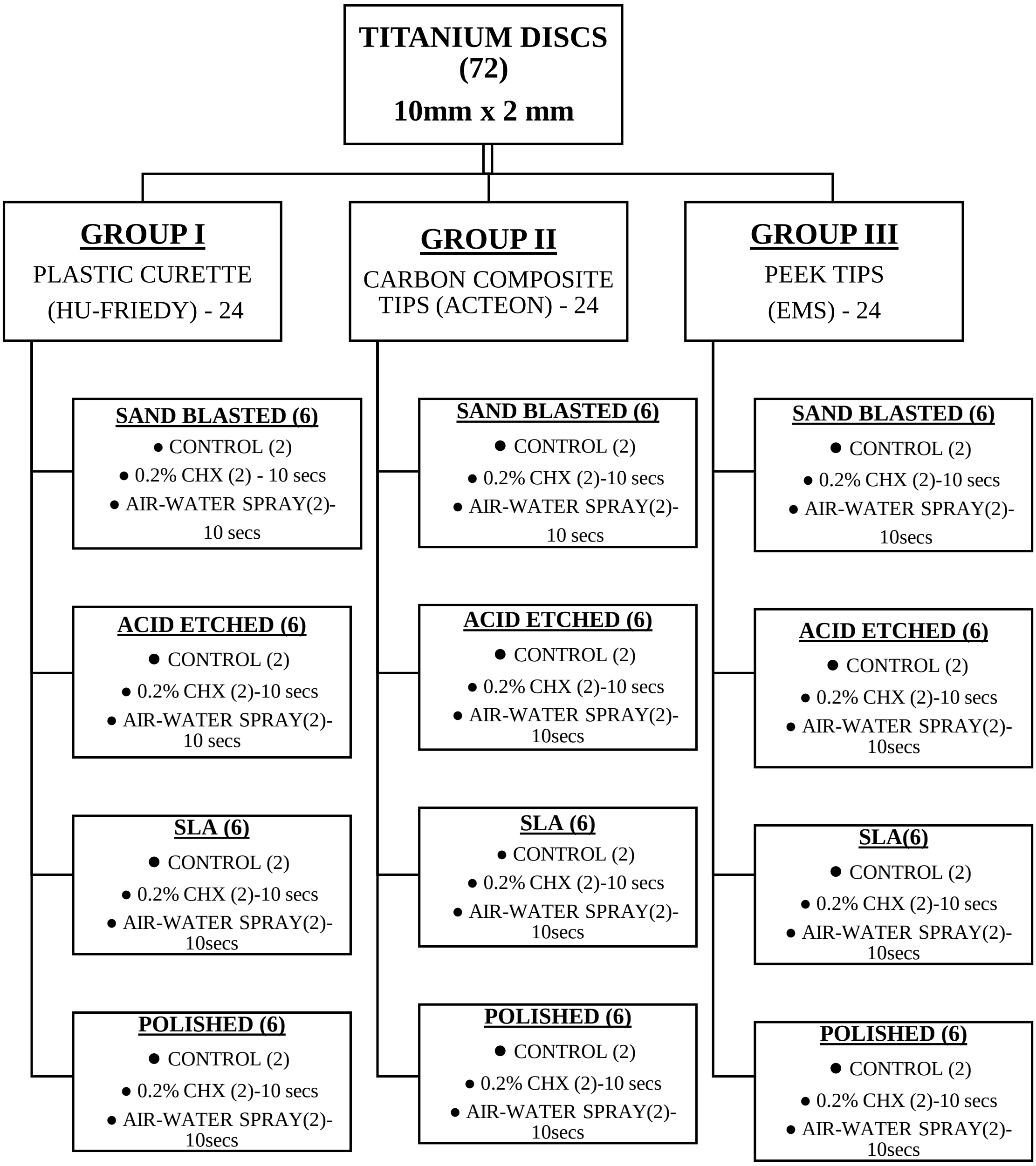
Grade 4 titanium discs were randomly divided into 5 groups and each group was divided into 2 subgroups, with one contaminated with *A. actinomycetemcomitans*, and the other contaminated with *P. gingivalis*. Group 1 did not receive bacterial inoculation or surface debridement and served as a control. Group 2 received *A. actinomycetemcomitans* or *P. gingivalis* inoculation, separately. Group 3 received bacterial inoculation and titanium curette debridement, followed by normal saline irrigation. Group 4 received bacterial inoculation, curette debridement, normal saline irrigation and ultrasonication. Group 5 received bacterial inoculation, curette debridement, normal saline irrigation and placement in 0.12% CHX. Results showed that after treatment, *A. actinomycetemcomitans* and *P. gingivalis* biofilms noticeably reduced surface hydrophilicity. Groups 3-5 showed decreased hydrophilicity and fewer adhered osteoblast cells compared with the control group. Although ultrasonication was more effective in removing LPS than curette debridement and CHX, cell adhesion was not as high as with clean titanium discs. Thus it was concluded that the non-surgical treatment used in this study was not effective in removing LPS from titanium surfaces and increasing osteoblast adhesion [50].

MATERIALS & METHODS

This in-vitro study was conducted at the Department of Periodontics, Sri Ramakrishna Dental College and Hospital, Coimbatore.

ARMAMENTARIUM:

1. Sandblasted titanium discs
2. Acid etched titanium discs
3. SLA treated titanium discs
4. Polished titanium discs
5. PEEK ultrasonic tip (EMS Piezon Systems)
6. Carbon composite ultrasonic tip (PH1 Satelec, Suprason)
7. Plastic curette (Columbia 4R/4L, Hu- Friedy)
8. Forceps
9. Air-water spray (3 way syringe)
10. 0.2% chlorhexidine mouthrinse
11. 2 ml disposable syringe
12. Cover slip
13. Confocal Laser Scanning Microscope (Carl Zeiss LSM 700)



Flowchart: Methodology

TITANIUM DISCS:

72 Titanium discs made of commercially pure Ti (ASTM Grade 4) measuring 10 mm in diameter and 2 mm in thickness were used in the study. The titanium discs were fabricated from Ti bars of 10 mm in diameter. The discs were fabricated at VR Industries, Ekkaduthangal, Chennai.

The discs were divided into 3 Groups of 24 discs each. They were instrumented using the following instruments:

Group I: A plastic curette (Columbia 4R/4L, Implacare, Hu- Friedy) which is made of Plasteel – a high grade unfilled resin.

Group II: A carbon composite tip on ultrasonic scaler A (PH1, Satelec, Suprason). These tips are made of fibre reinforced plastic containing carbon fibres.

Group III: A plastic tip on ultrasonic scaler B (Polyetheretherketone tip-PEEK, EMS Piezon Systems). These tips are made of polyetheretherketone (PEEK) - a thermoplastic polymer.

Ultrasonic scaler A was applied at power setting 3 at 25 to 32 kHz and ultrasonic scaler B was applied at a power setting of 3 at 27 to 33 kHz according to the manufacturer's manual.

The discs were surface modified to mimic surface topography of commercially available dental implants. The following surface modifications were done:

- Sand blasting
- Acid etching
- Sand blasting and acid etching (SLA)
- Polishing

Out of the 24 discs in each Group, 6 discs were sand blasted, 6 were acid etched, 6 were SLA treated and 6 were polished.

All the discs were polished using # 800 grit silicon carbide metallographic papers, washed in distilled water, cleaned and dried at room temperature. These discs were then subjected to the following surface modifications:

Sand blasting: Sand blasting was done on one side of the disc, with 250 µm alumina particles at 20 psi for 1 minute with a fixed distance of 1 cm between the sample and blasting tip.

Acid etching: Discs were acid etched by boiling in 5% sulphuric acid for 15 hours at 60°c.

SLA: Discs were sand blasted with 250 µm alumina particles followed by chemical treatment in boiling 5% sulphuric acid for 15 hours at 60°c.

Polishing: Discs were manually polished using # 800 - # 2000 grit silicon carbide metallographic papers.

INSTRUMENTATION OF DISCS:

- The discs were instrumented by vertical 40 strokes and 40 horizontal strokes.
- The scaler tips and the plastic curette were angulated tangentially, and care was taken to place minimal lateral pressure on the titanium.

EFFICACY OF REMOVAL OF PLASTIC REMNANTS:

From each instrument group of 6 discs, 2 discs each were treated with:

1. Air-water spray from a three way syringe for 10 seconds
2. Irrigated with 0.2 % chlorhexidine mouthwash using a 2 ml syringe for 10 seconds.
3. No irrigation which served as controls

The discs were then dried in open air for 1 hour.

EVALUATION OF THE SURFACE AREA OF PLASTIC REMNANTS AFTER INSTRUMENTATION:

An image was taken of the centre of each disc with a Confocal Laser Scanning Microscope (Carl Zeiss LSM 700) equipped with a 488nm argon laser using a X20 Plan – Apochromat objective lens. Images sized 461.2µm X 461.2µm were captured and digitized.

A wavelength of 488 nm was used to capture the images in green. MATLAB (version R2009b, The MathWorks Inc., USA), a digital image analysis software was used to quantify the area with auto fluorescence. The surface area was calculated in percentage. The evaluation using Confocal Laser Scanning Microscope was done at the National Facility for Clinical Trials, Interdisciplinary Institute of Indian System of Medicine (IIISM), SRM University, Kattankalathur, Chennai.

FIGURES

Figure 1: Group I - Plastic Curette (Hu-Friedy)

Figure 2: Group II – Carbon Composite Tip (Satelec)

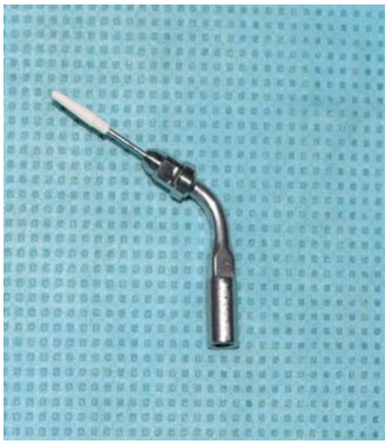


Figure 3: Group III – Peek Tip (EMS)

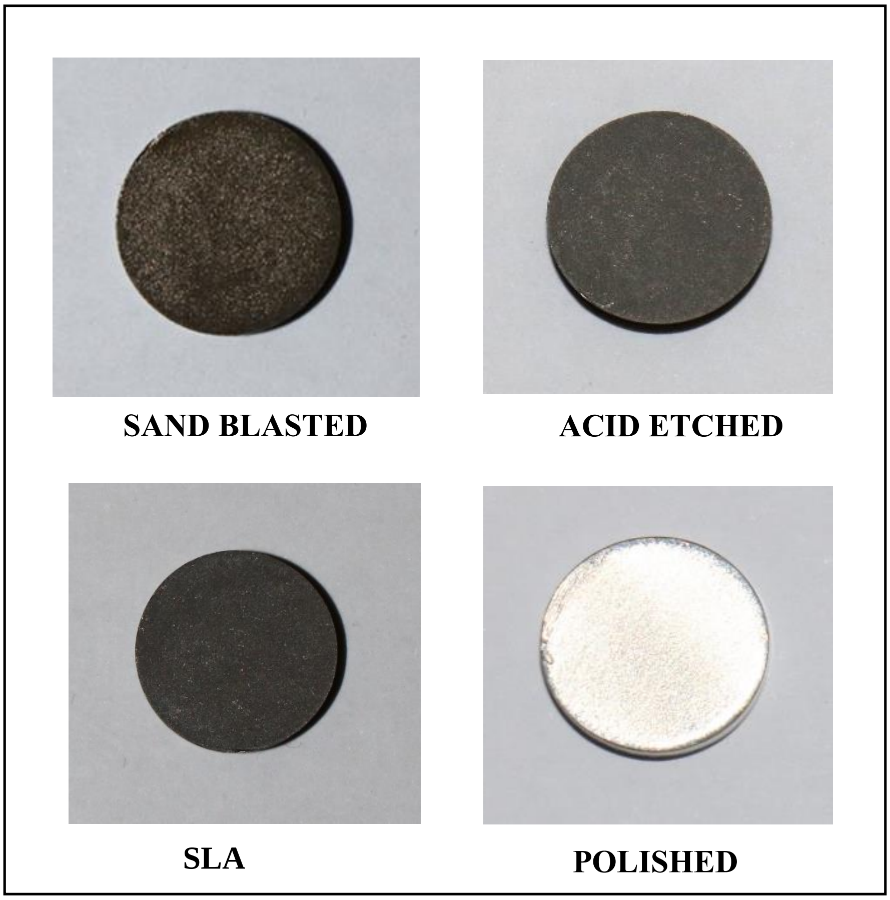


Figure 4: Surface modified titanium discs before instrumentation

Figure 5: Instrumentation using Plastic curette (Hu-Friedy)

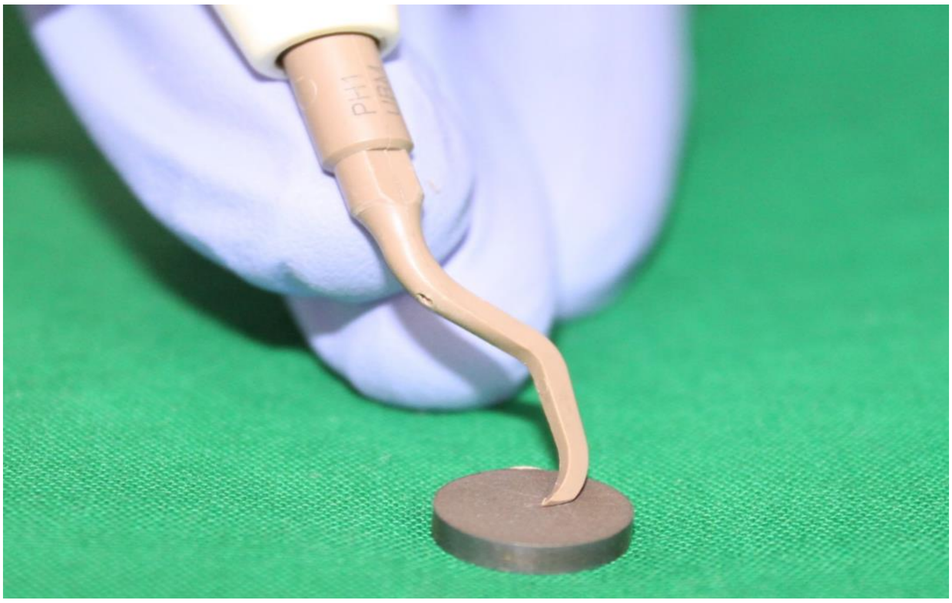


Figure 6: Instrumentation using Carbon composite tip (Satelec)

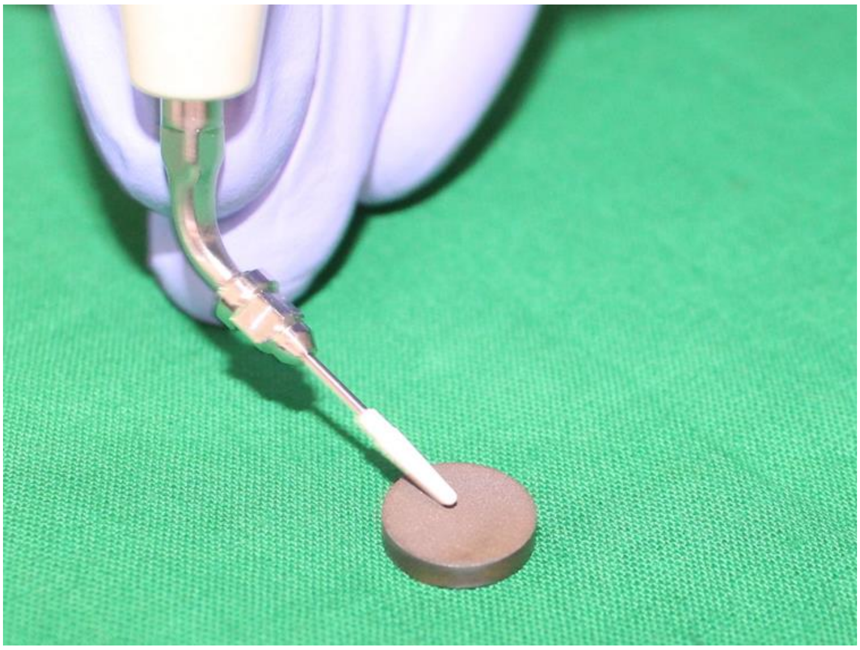


Figure 7: Instrumentation using PEEK tip (EMS)

Figure 8: Irrigation using 0.2% chlorhexidine

Figure 9: Irrigation using air-water spray

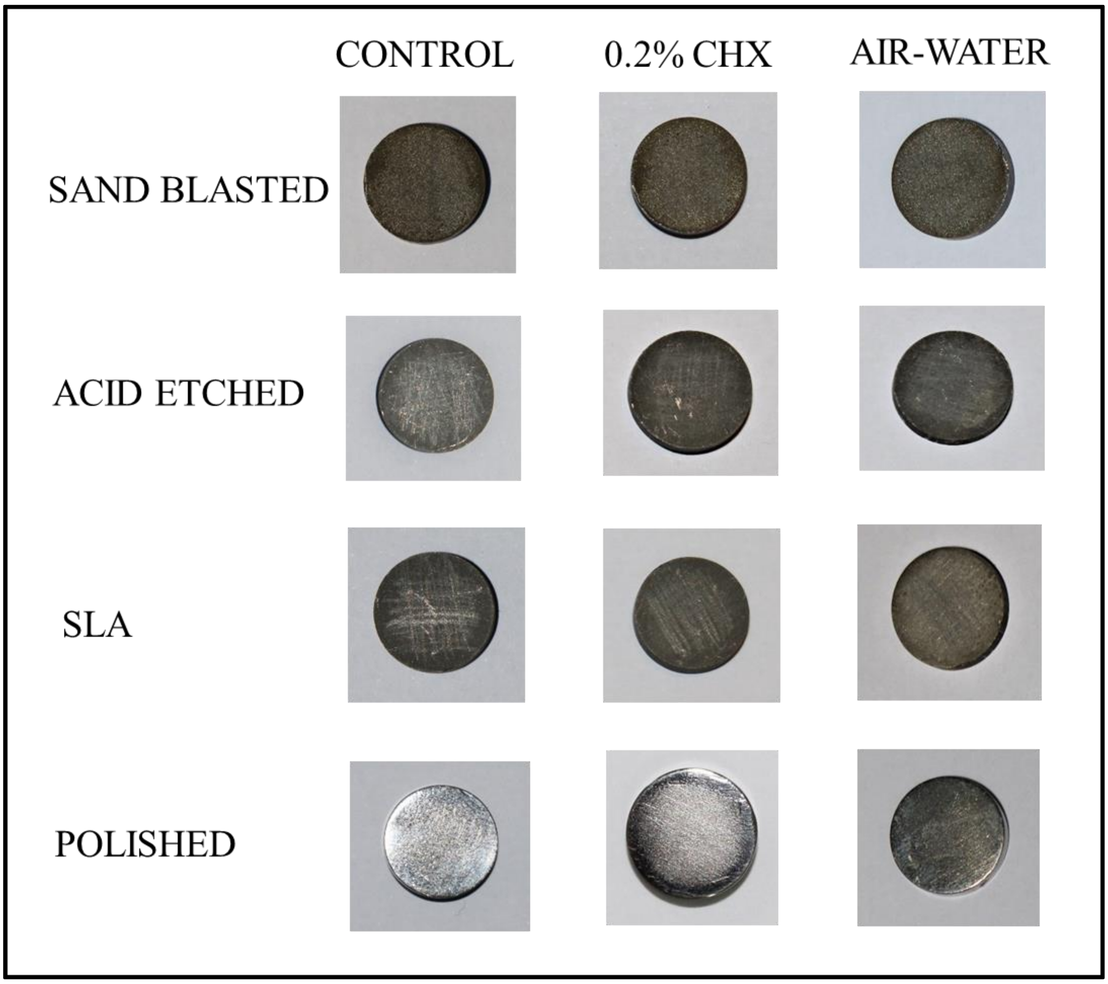


Figure 10: Surface modified titanium discs following instrumentation and irrigation with Group I – Plastic curette

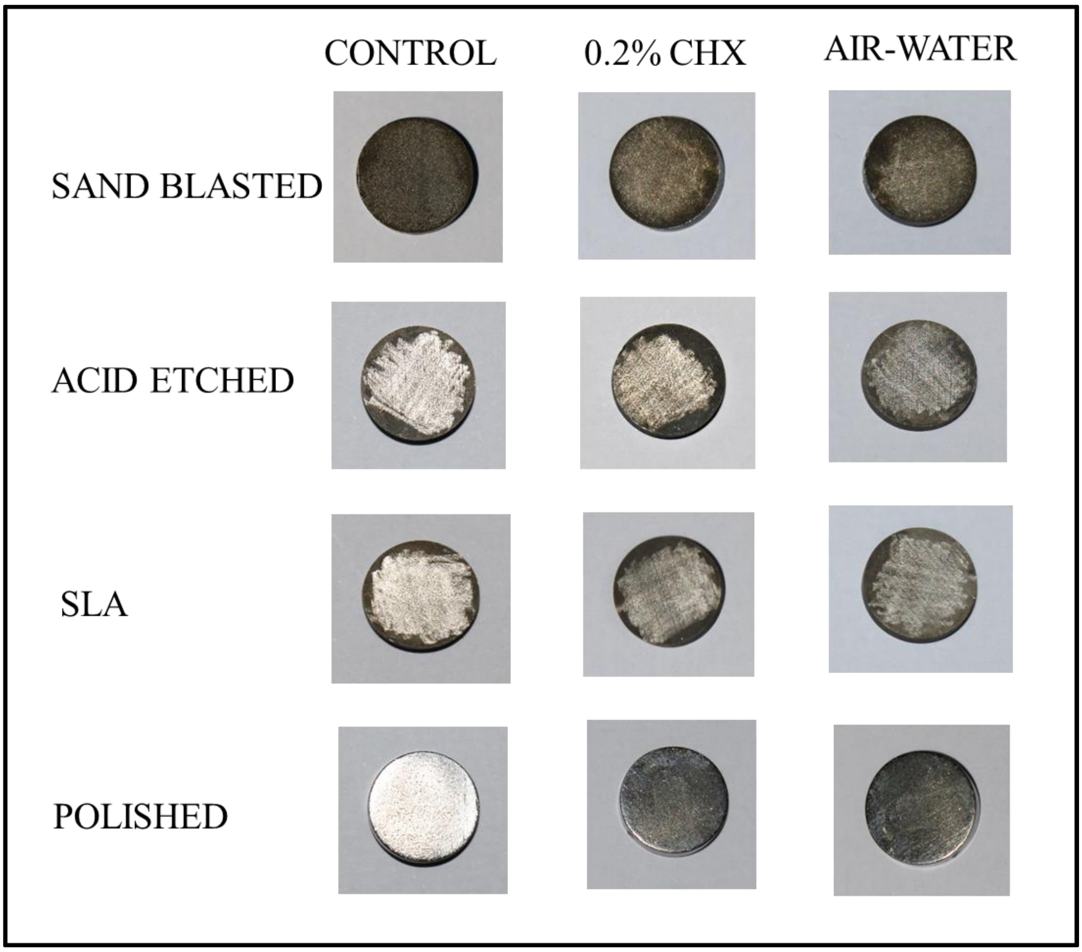


Figure 11: Surface modified titanium discs following instrumentation and irrigation with Group II – Carbon composite tip

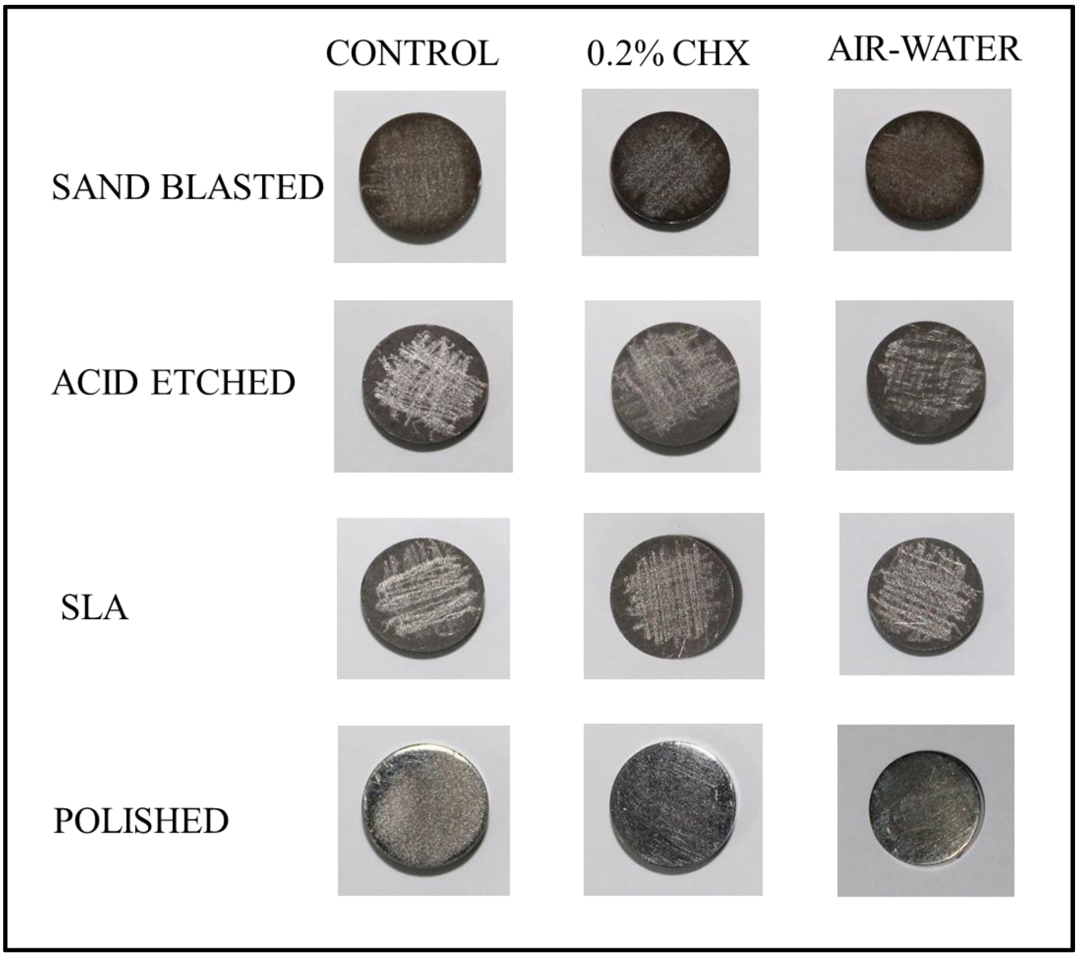


Figure 12: Surface modified titanium discs following instrumentation and irrigation with Group III- PEEK tip

Figure 13: Confocal laser scanning microscope

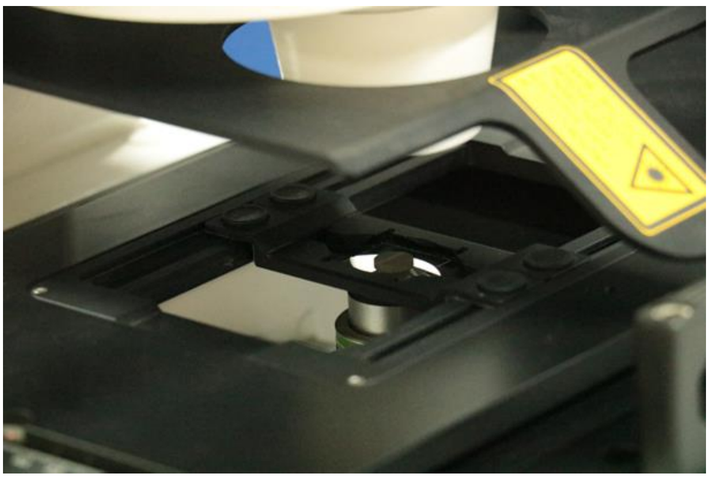


Figure 14: Titanium disc in focus under confocal laser scanning microscope

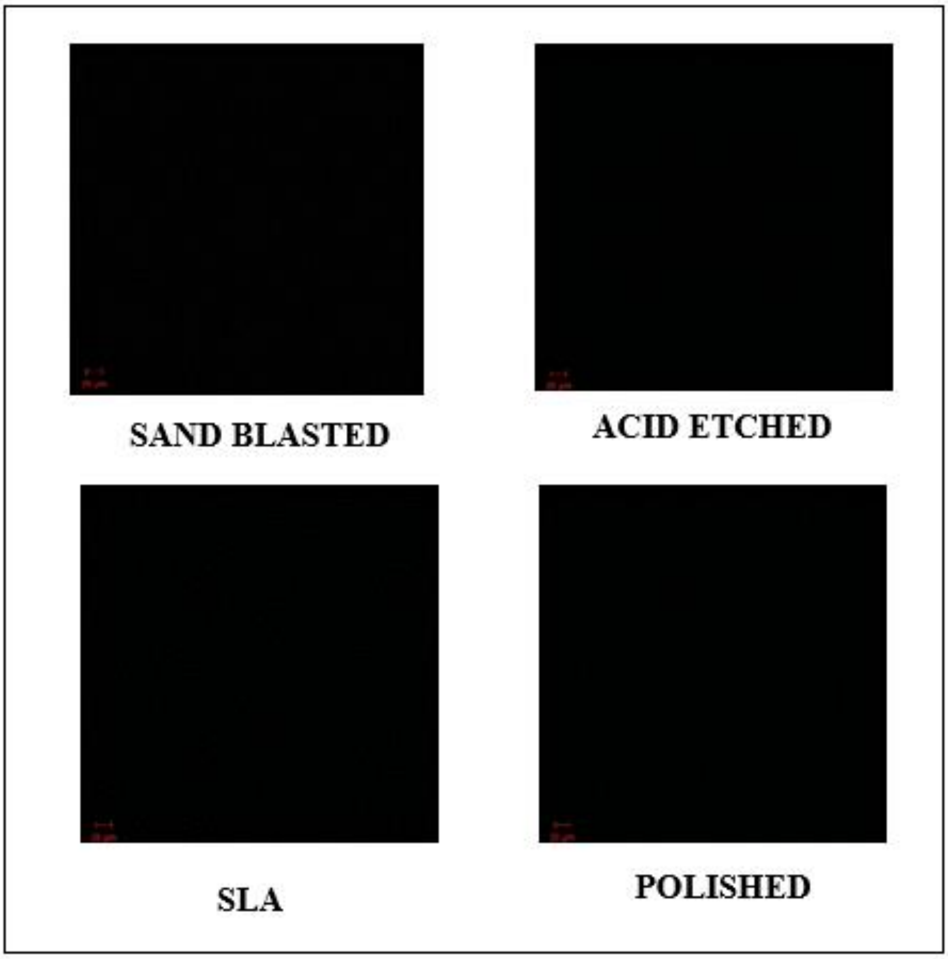


Figure 15: Surface modified titanium discs before instrumentation under confocal laser scanning microscope

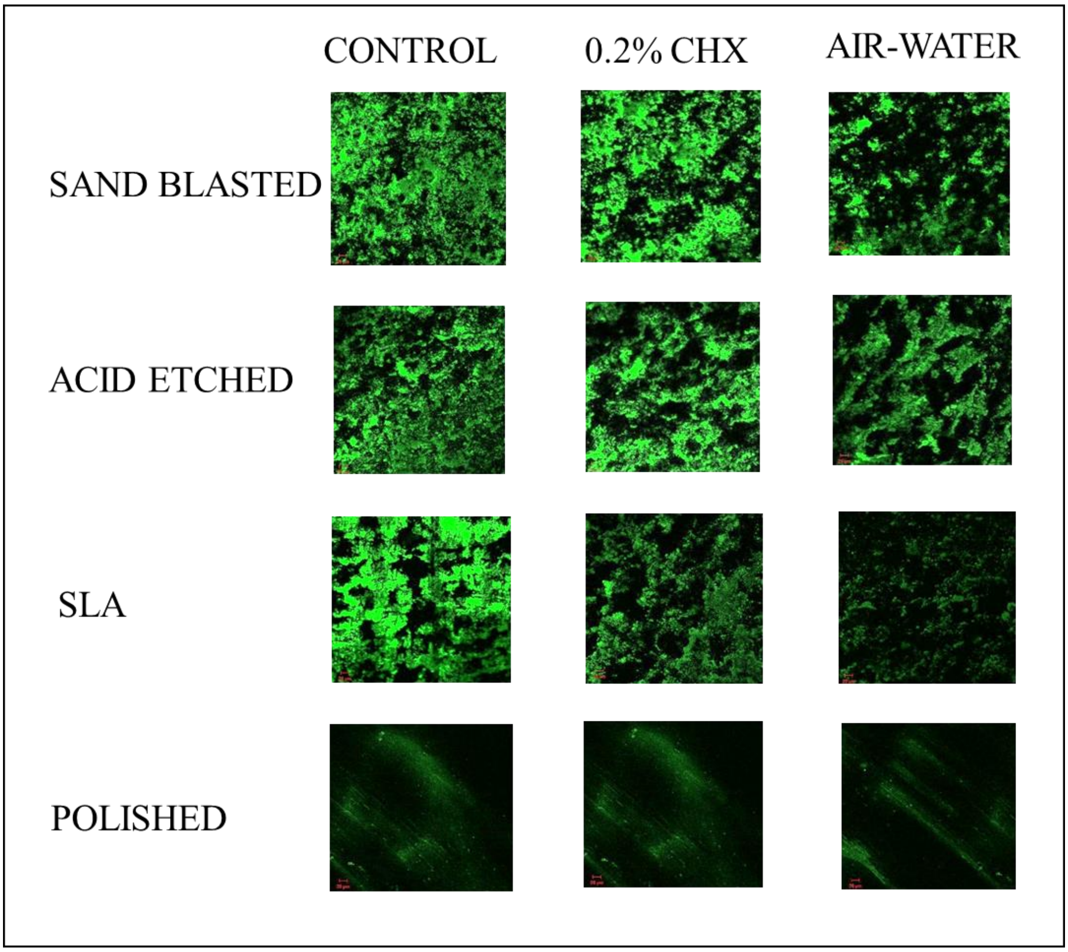


Figure 16: Surface modified titanium discs following instrumentation and irrigation with Group I – Plastic curette (Hu-Friedy) under confocal laser scanning microscope

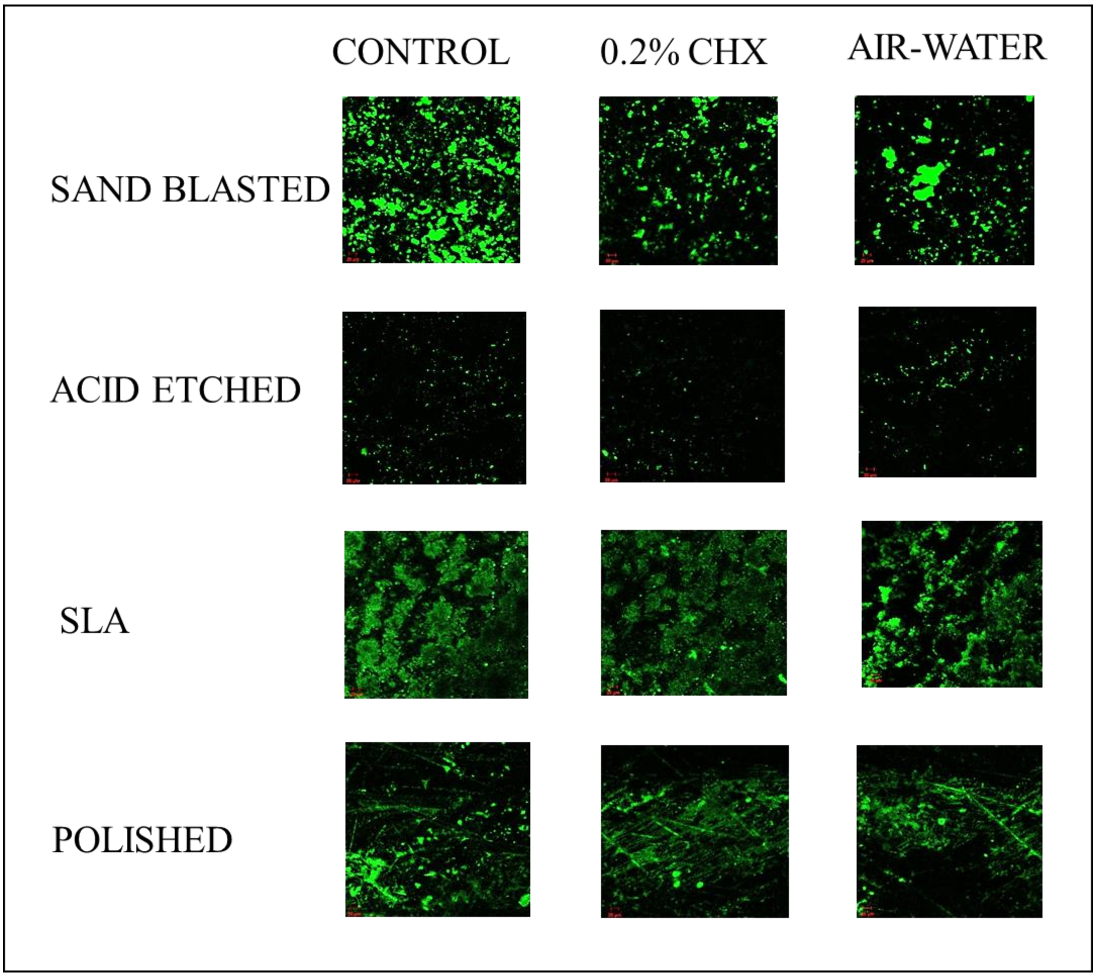


Figure 17: Surface modified titanium discs following instrumentation and irrigation with Group II – Carbon composite tip (Satelec) under confocal laser scanning microscope

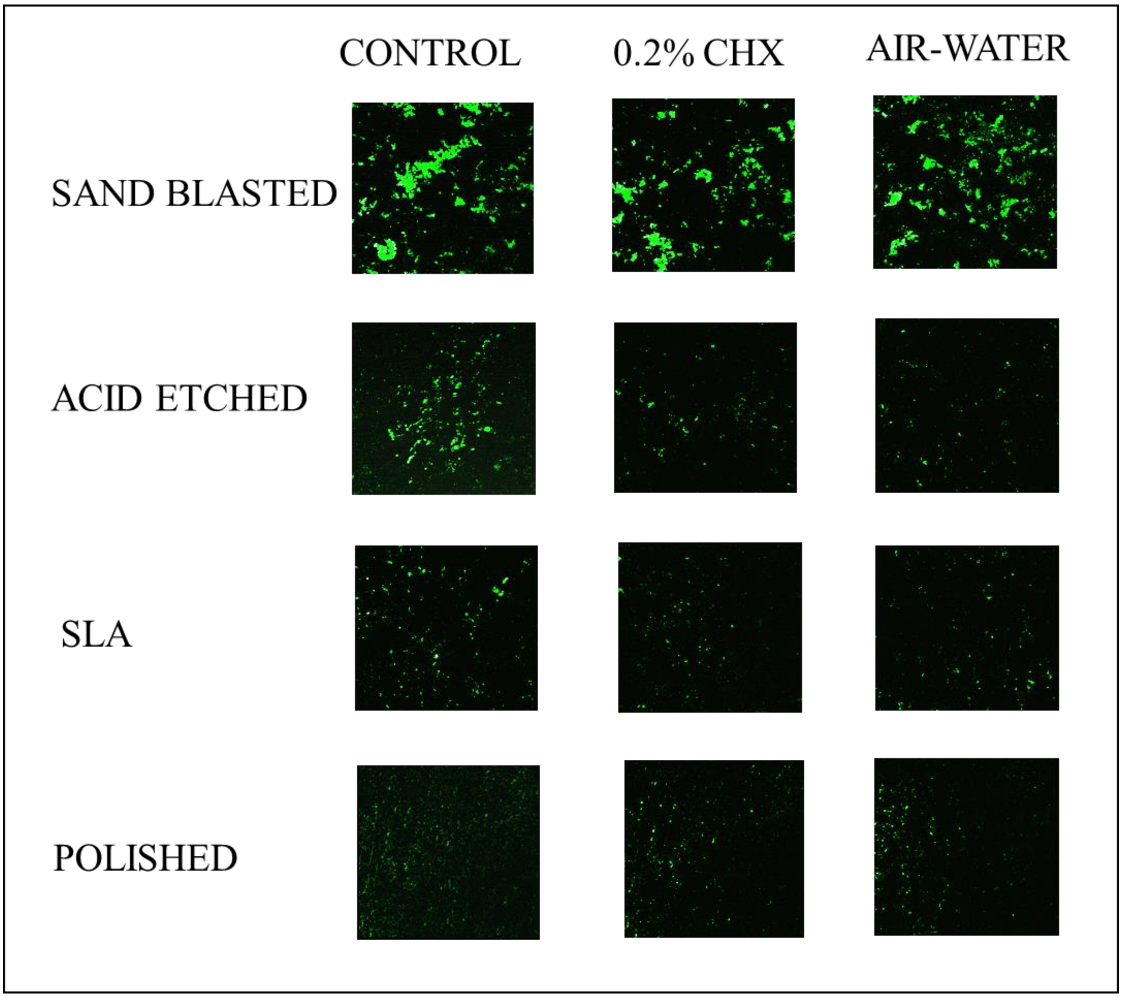


Figure 18: Surface modified titanium discs following instrumentation and irrigation with Group III- PEEK tip (EMS) under confocal laser scanning microscope

RESULTS

STATISTICAL ANALYSIS:

In this study, the mean areas and standard deviations were calculated for each group and subgroup. Statistical analysis was performed using One way ANOVA followed by Tukeys Post HOC test (SPSS version 16 for Windows, SPSS Inc.). The statistical significance of any differences was evaluated, with significance set at $p < 0.05$.

72 titanium discs were divided into 3 groups of 24 discs each. The discs were instrumented using the following:

GROUP I - Plastic curette (Hu- Friedy)

GROUP II - Carbon composite tip (Satelec)

GROUP III - PEEK tip (EMS)

MEAN AREA COVERED BY PLASTIC REMNANTS FOLLOWING INSTRUMENTATION:

After instrumentation, the surface area occupied by the plastic remnants was calculated in % using the MATLAB software.

Following instrumentation on titanium discs, it was found that Group I (Plastic curette- Hu Friedy) left behind the maximum plastic debris with a mean area of 47.38 ± 1.26% followed by Group II (Carbon composite tip – Satelec) with a mean area of 38.72 ± 1.03%. The least amount of plastic remnants were found with Group III (PEEK Tip - EMS) which had a mean area of 27.4 ± 7.49% (Table 1, Graph 1)[#].

On comparison, it was found that Group I left more plastic debris when

[#]All tables and graphs at the end of the result section compared to Group II (p = 0.017*) and Group III (p = 0.000*) which was found to be statistically significant. It was also found that Group II left more debris when compared to Group III which was found to be statistically significant (p=0.005*) (Table 2).

MEAN AREA COVERED BY PLASTIC REMNANTS AFTER INSTRUMENTATION AND IRRIGATION:

Following instrumentation, 10 seconds of air- water spray or irrigation with 0.2 % chlorhexidine using a 2ml plastic syringe was used to remove the plastic remnants. Irrigation with the above mentioned methods led to a varying degree of decrease in the amount of remnants left on all the discs in all the groups.

In Group I, the use of air-water spray decreased the surface area covered by plastic debris from 47.38 ± 1.26% (control) to 35.22 ± 8.65% and the use of 0.2% chlorhexidine irrigation reduced it to 42.76 ± 1.02% (Table 3, Graph 2).

In Group II, the use of air-water spray decreased the surface area covered by plastic debris from 38.72 ± 1.03% (control) to 27.58 ± 7.94% and the use of 0.2% chlorhexidine irrigation reduced it to 34.95 ± 1.01% (Table 4, Graph 3).

In Group III, the use of air-water spray decreased the surface area covered by plastic debris from 27.41 ± 7.49% (control) to 20.97 ± 5.71% and the use of 0.2% chlorhexidine irrigation reduced it to 25.16 ± 8.03% (Table 5, Graph 4).

MEAN AREA COVERED BY PLASTIC REMNANTS AFTER INSTRUMENTATION ON VARIOUS MODIFIED SURFACES FOLLOWED BY IRRIGATION:

In Group I,

- In sandblasted titanium discs, it was found that the mean area covered by plastic remnants reduced from 62.73 ± 6.08% to 52.85 ± 3.37% after 0.2%

chlorhexidine irrigation and to 47.29 ± 1.16 % after irrigation with air-water spray (Table 6, Graph 5).

- In acid etched titanium discs, it was found that the mean area covered by plastic remnants reduced from 44.72 ± 3.19 to 40.14 ± 1.63% after 0.2% chlorhexidine irrigation and to 35.79 ± 3.64 % after irrigation with air-water spray (Table 6, Graph 6).
- In SLA titanium discs, it was found that the mean area covered by plastic remnants reduced from 51.09 ± 0.88% to 49.64 ± 1.3% after 0.2% chlorhexidine irrigation and to 32.65 ± 0.80% after irrigation with air-water spray (Table 6, Graph 7).
- In polished titanium discs, it was found that the mean area covered by plastic remnants reduced from 30.97± 4.95% to 28.42 ± 0.4% after 0.2% chlorhexidine irrigation and to 25.14 ± 1.14 % after irrigation with air-water spray (Table 6, Graph 8).

In Group II,

- In sandblasted titanium discs, it was found that the mean area covered by plastic remnants reduced from 49.54 ± 2.48% to 44.46 ± 1.78% after 0.2% chlorhexidine irrigation and to 30.87 ± 2.06 % after irrigation with air-water spray (Table 7, Graph 9).
- In acid etched titanium discs, it was found that the mean area covered by plastic remnants reduced from 35.57 ± 2.05% to 32.97 ± 2.20% after 0.2% chlorhexidine irrigation and to 28.52 ± 1.54 % after irrigation with air-water spray (Table 7, Graph 10).
- In SLA titanium discs, it was found that the mean area covered by plastic remnants reduced from 45.13 ± 1.43% to 42.04 ± 1.09% after 0.2%

chlorhexidine irrigation and to 35.27 ± 1.84% after irrigation with air-water spray (Table 7, Graph 11).

- In polished titanium discs, it was found that the mean area covered by plastic remnants reduced from 24.65 ± 1.99% to 20.33 ± 1.08% after 0.2% chlorhexidine irrigation and to 15.65 ± 2.42 % after irrigation with air-water spray (Table 7, Graph 12).

In Group III,

- In sandblasted titanium discs, it was found that the mean area covered by plastic remnants reduced from 34.75 ± 2.56% to 32.50 ± 1.77% after 0.2% chlorhexidine irrigation and to 23.79 ± 0.46 % after irrigation with air-water spray (Table 8, Graph 13).
- In acid etched titanium discs, it was found that the mean area covered by plastic remnants reduced from 28.29 ± 1.58% to 26.17 ± 1.83% after 0.2% chlorhexidine irrigation and to 22.87 ± 0.77 % after irrigation with air-water spray (Table 8, Graph 14).
- In SLA titanium discs, it was found that the mean area covered by plastic remnants reduced from 30.46 ± 1.47% to 29.12 ± 1.38% after 0.2% chlorhexidine irrigation and to 25.09 ± 1.76% after irrigation with air-water spray (Table 8, Graph 15).
- In polished titanium discs, it was found that the mean area covered by plastic remnants reduced from 16.14± 0.59% to 12.87 ± 0.77% after 0.2% chlorhexidine irrigation and to 12.14 ± 3.43 % after irrigation with air-water spray (Table 8, Graph 16).

On comparing the effect of irrigation on various surface modifications, it was found that irrigation with 0.2% chlorhexidine seemed to still leave behind statistically significant amount of plastic debris on sandblasted group compared to polished surfaces (p = 0.001*). Significant amount of plastic debris was also found in SLA surface when compared to polished (p = 0.002*) (Table 9).

Following irrigation with air –water spray, it was found that sand blasted surfaces retained significant amount of plastic debris when compared to polished surfaces (p =0.005*). Significant amount of plastic debris was found in SLA surfaces when compared to polished (p=0.025*) (Table 10).

MEAN AREA COVERED BY PLASTIC REMNANTS IN 3 GROUPS AFTER INSTRUMENTATION AND IRRIGATION:

On irrigation with 0.2% chlorhexidine, it was found that Group I retained a mean area of 42.76 ± 1.02 % of plastic remnants. Group II had a mean area of 34.95 ± 1.01% and Group III had a mean area of 25.16 ± 8.03% (Table 11, Graph 17). It was found that Group I retained more plastic remnants as compared to Group III and the difference was found to be statistically significant (p = 0.004*).

On irrigation with the air-water spray, it was found that Group I retained a mean area of 35.22 ± 8.6 % of plastic remnants. Group II retained a mean area of 27.58 ± 7.9% and Group III had retained a mean area of 20.97 ± 5.71% (Table 12, Graph 18). It was found that Group I retained more plastic debris as compared to Group III and the difference was found to be statistically significant (p =0.003*).

Overall, it was found that plastic remnants remained after instrumentation, regardless of the irrigation method used. But it was found that the air-water spray had a better irrigating effect in removing the plastic remnants after instrumentation when compared to 0.2% chlorhexidine irrigation.

Table 1: MEAN AREA COVERED BY PLASTIC REMNANTS AFTER INSTRUMENTATION WITH 3 GROUPS

Instruments	**N**	**Minimum (Area in %)**	**Maximum (Area in %)**	**Mean (Area in %)**	**Std. Deviation**
GROUP I	24	24.34	67.04	47.3834	1.2672
GROUP II	24	13.94	51.30	38.7276	1.0339
GROUP III	24	9.71	36.56	27.4136	7.4994

Table 2: COMPARSION AMONG THE GROUPS

GROUPS		**p VALUE**
GROUP I	GROUP II	**0.017***
	GROUP III	**0.000***
GROUP II	GROUP III	**0.005***

Note: * denotes significance of ($p<0.05$)

Table 3: MEAN AREA COVERED BY PLASTIC REMNANTS AFTER INSTRUMENTATION AND IRRIGATION IN GROUP I

GROUP I	**Irrigation**	**Mean (Area In %)**	**Standard Deviation (Area In %)**
	AIR-WATER	35.2206	8.65865
	CHX	42.7617	1.0277

Table 4: MEAN AREA COVERED BY PLASTIC REMNANTS AFTER INSTRUMENTATION AND IRRIGATION IN GROUP II

GROUP II	**Irrigation**	**Mean (Area In %)**	**Standard Deviation (Area In %)**
	AIR-WATER	27.5836	7.949
	CHX	34.9561	1.0189

Table 5: MEAN AREA COVERED BY PLASTIC REMNANTS AFTER INSTRUMENTATION AND IRRIGATION IN GROUP III

GROUP III	**Irrigation**	**Mean (Area In %)**	**Standard Deviation (Area In %)**
	AIR-WATER	20.9758	5.7174
	CHX	25.1685	8.0393

Table 6: MEAN AREA COVERED BY PLASTIC REMNANTS AFTER INSTRUMENTATION ON VARIOUS MODIFIED SURFACES FOLLOWED BY IRRIGATION IN GROUP I

Group I (Plastic Curette- Hu Friedy)		**N**	**Mean (Area In %)**	**Standard Deviation (Area In %)**
SAND BLASTED	CONTROL	2	62.7337	6.08345
	CHX	2	52.8595	3.37785
	AIR-WATER	2	47.2917	1.16192
ACID ETCHED	CONTROL	2	44.7291	3.19188
	CHX	2	40.1402	1.63752
	AIR-WATER	2	35.7920	3.64570
SLA	CONTROL	2	51.0989	0.88643
	CHX	2	49.6441	1.30546
	AIR-WATER	2	32.6518	0.80002
POLISHED	CONTROL	2	30.9721	4.95116
	CHX	2	28.4271	0.40305
	AIR-WATER	2	25.1471	1.14000

Table 7: MEAN AREA COVERED BY PLASTIC REMNANTS AFTER INSTRUMENTATION ON VARIOUS MODIFIED SURFACES FOLLOWED BY IRRIGATION IN GROUP II

Group II (Carbon Composite Tip- Acteon)		**N**	**Mean (Area In %)**	**Standard Deviation (Area In %)**
SAND BLASTED	CONTROL	2	49.5412	2.48930
	CHX	2	44.4622	1.78898
	AIR-WATER	2	30.8788	2.06701
ACID ETCHED	CONTROL	2	35.5721	2.05146
	CHX	2	32.9781	2.20207
	AIR-WATER	2	28.5211	1.54701
SLA	CONTROL	2	45.1390	1.43274
	CHX	2	42.0446	1.09319
	AIR-WATER	2	35.2780	1.84682
POLISHED	CONTROL	2	24.6582	1.99560
	CHX	2	20.3396	1.08640
	AIR-WATER	2	15.6563	2.42424

Table 8: MEAN AREA COVERED BY PLASTIC REMNANTS AFTER INSTRUMENTATION ON VARIOUS MODIFIED SURFACES FOLLOWED BY IRRIGATION IN GROUP III

Group III (PEEK Tip - EMS)		**N**	**Mean (Area In %)**	**Standard Deviation (Area In %)**
SAND BLASTED	CONTROL	2	34.7521	2.56128
	CHX	2	32.5041	1.77654
	AIR-WATER	2	23.7911	0.46711
ACID ETCHED	CONTROL	2	28.2951	1.58392
	CHX	2	26.1742	1.83551
	AIR-WATER	2	22.8715	0.77174
SLA	CONTROL	2	30.4672	1.47884
	CHX	2	29.1243	1.38197
	AIR-WATER	2	25.0985	1.76367
POLISHED	CONTROL	2	16.1402	0.59227
	CHX	2	12.8714	0.77796
	AIR-WATER	2	12.1421	3.43527

Table 9: <u>INTERGROUP COMPARSION AMONG VARIOUS MODIFIED SURFACES AFTER IRRIGATION WITH 0.2% CHX</u>

GROUPS		**p VALUE**
SANDBLASTED	POLISHED	**0.001***
SLA	POLISHED	**0.002***

Note: * denotes significance of (p<0.05)

Table 10: <u>INTERGROUP COMPARSION AMONG VARIOUS MODIFIED SURFACES AFTER IRRIGATION WITH AIR-WATER SPRAY</u>

GROUPS		**p VALUE**
SANDBLASTED	POLISHED	**0.005***
SLA	POLISHED	**0.025***

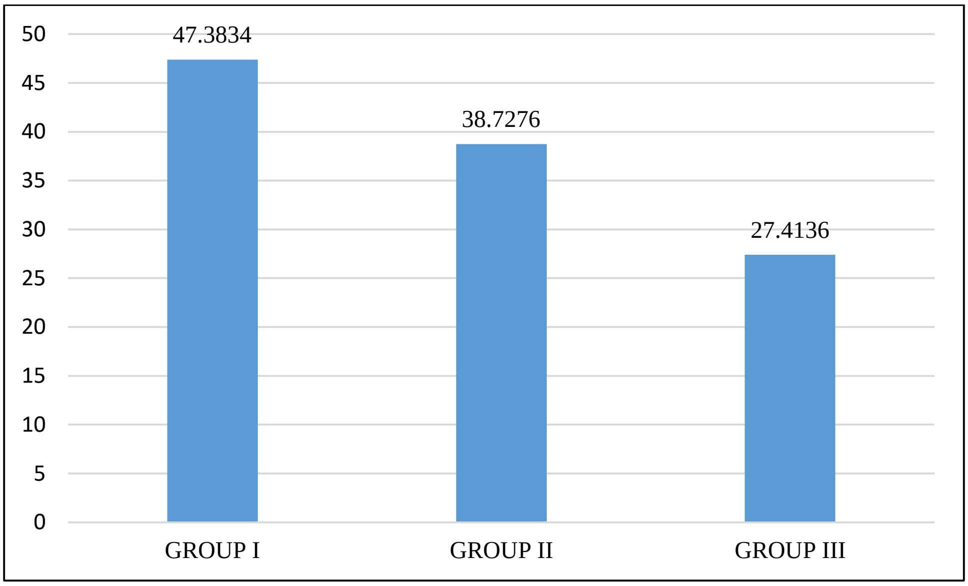
Note: * denotes significance of (p<0.05)

Table 11: MEAN AREA COVERED BY PLASTIC REMNANTS IN 3 GROUPS AFTER INSTRUMENTATION AND IRRIGATION WITH 0.2% CHX

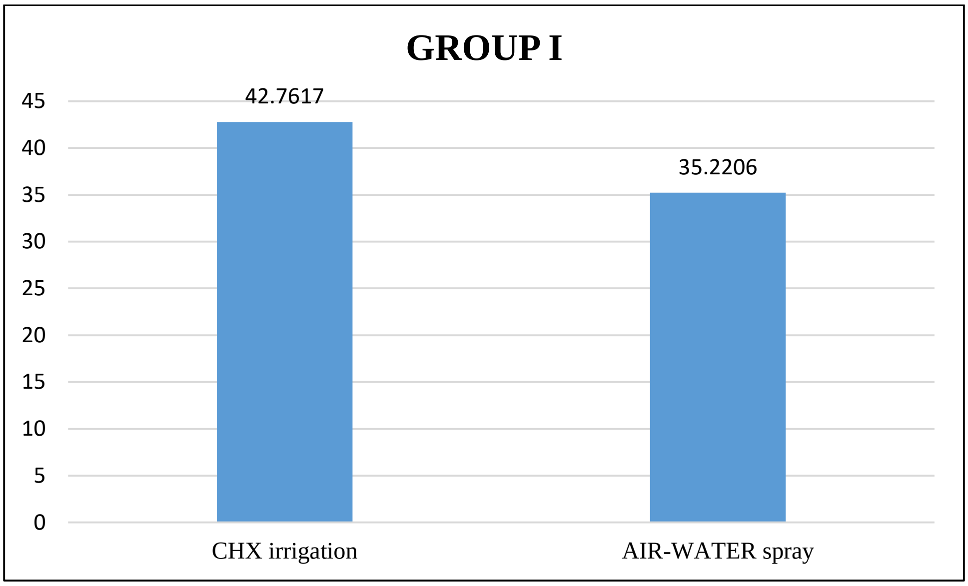
Instruments	**N**	**Mean (Area in %)**	**Std. Deviation (Area in %)**
GROUP I	24	42.7677	1.0277
GROUP II	24	34.9561	1.0189
GROUP III	24	25.1685	8.03936

Table 12: MEAN AREA COVERED BY PLASTIC REMNANTS IN 3 GROUPS AFTER INSTRUMENTATION AND IRRIGATION WITH AIR-WATER SPRAY

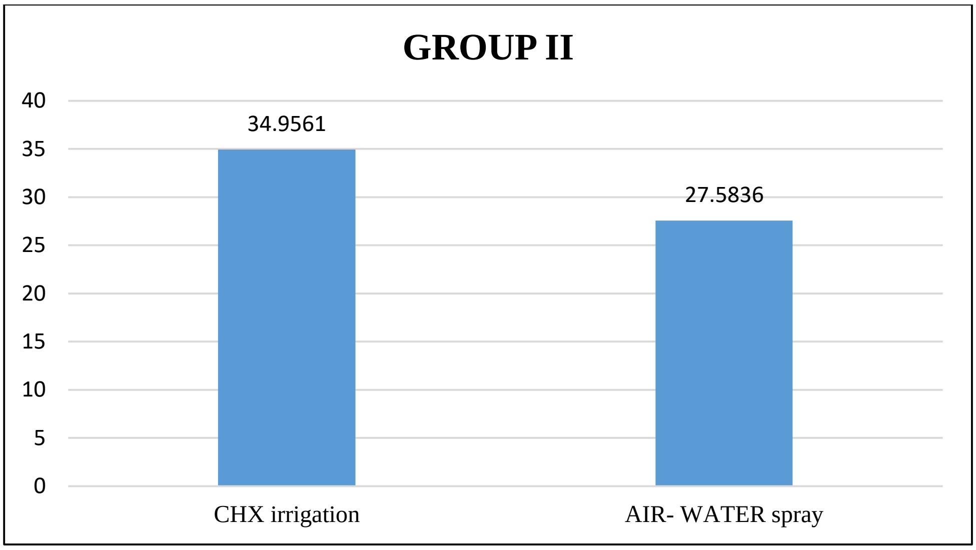
Instruments	**N**	**Mean (Area in %)**	**Std. Deviation (Area in %)**
GROUP I	24	35.2206	8.6586
GROUP II	24	27.5836	7.9494
GROUP III	24	20.9758	5.7174



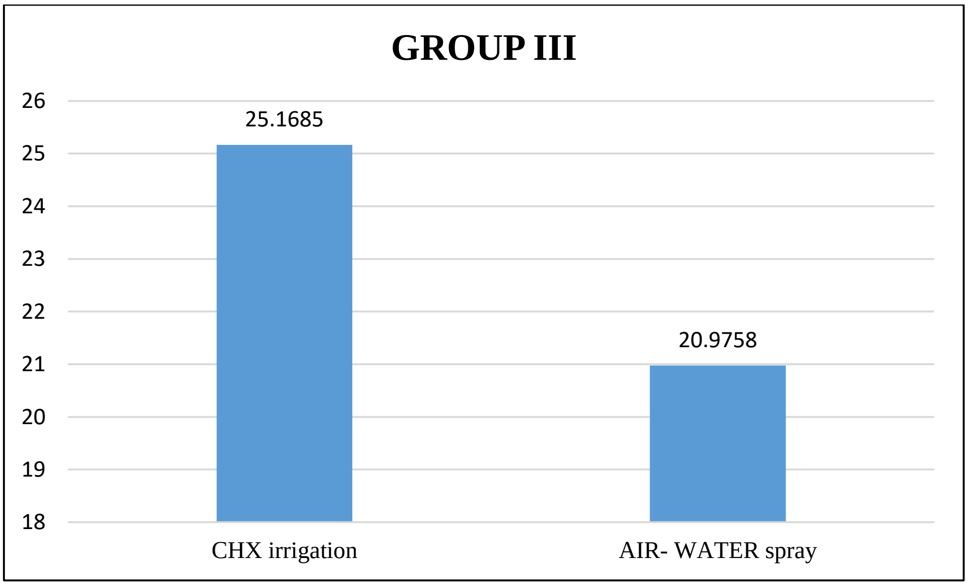
Graph 1: Mean area covered by plastic remnants after instrumentation in 3 Groups



Graph 2: Mean area covered by plastic remnants after irrigation with CHX and air-water spray in Group I

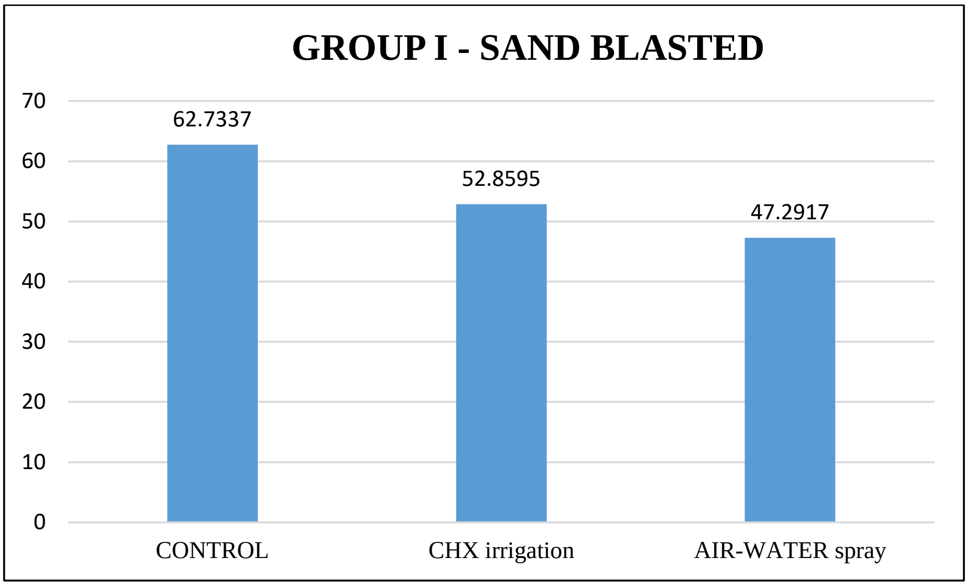


Graph 3: Mean area covered by plastic remnants after irrigation with CHX and air-water spray in Group II

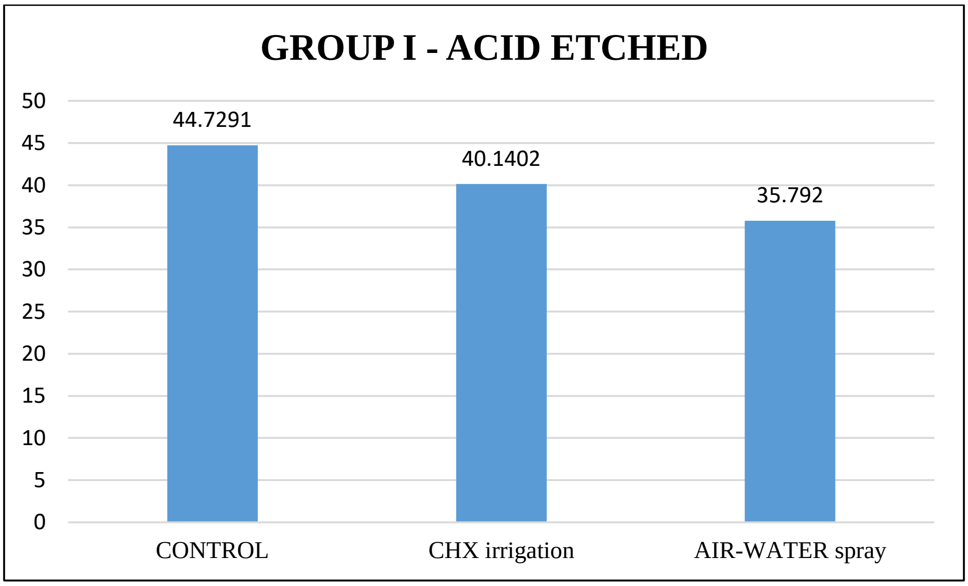


Graph 4: Mean area covered by plastic remnants after irrigation with CHX and air-water spray in Group III

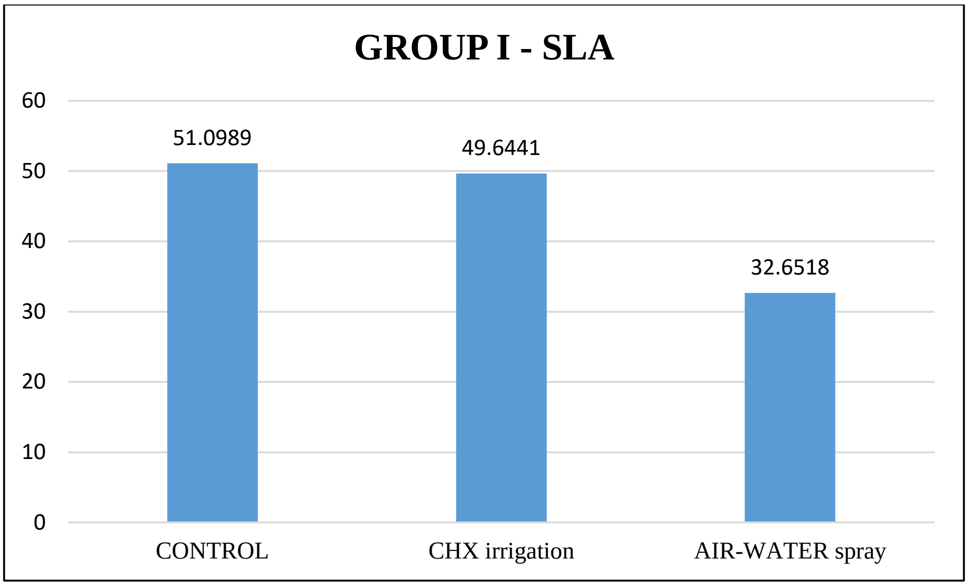
MEAN AREA OF PLASTIC REMNANTS AFTER INSTRUMENTATION AND IRRIGATION IN GROUP I



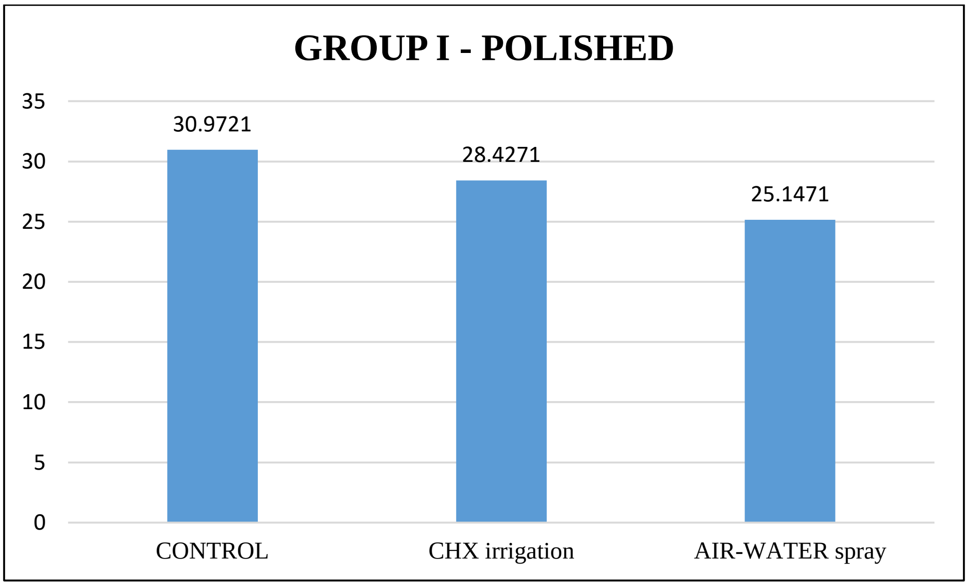
Graph 5: Mean area covered by plastic remnants after instrumentation and irrigation in Group I sand blasted titanium discs



Graph 6: Mean area covered by plastic remnants after instrumentation and irrigation in Group I acid etched titanium discs

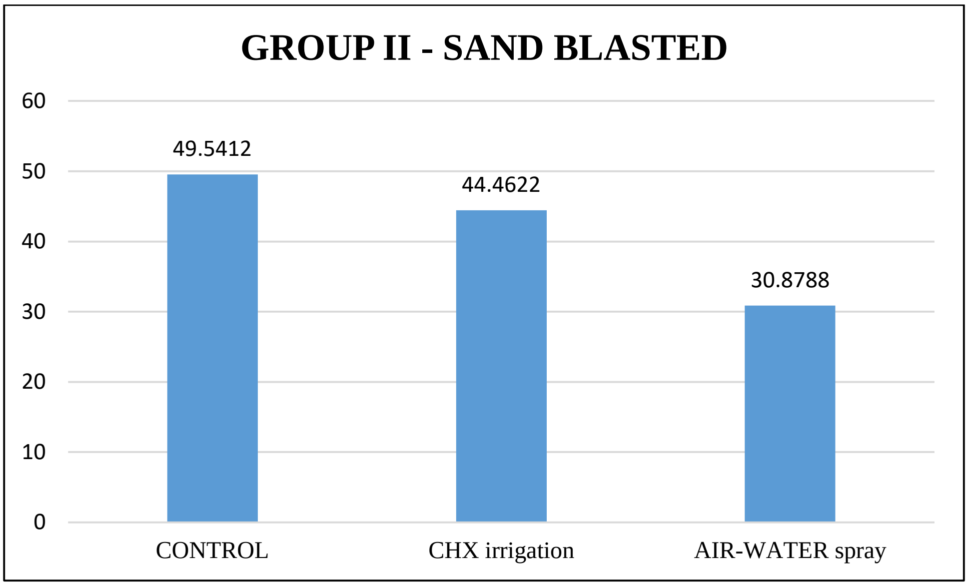


Graph 7: Mean area covered by plastic remnants after instrumentation and irrigation in Group I SLA titanium discs

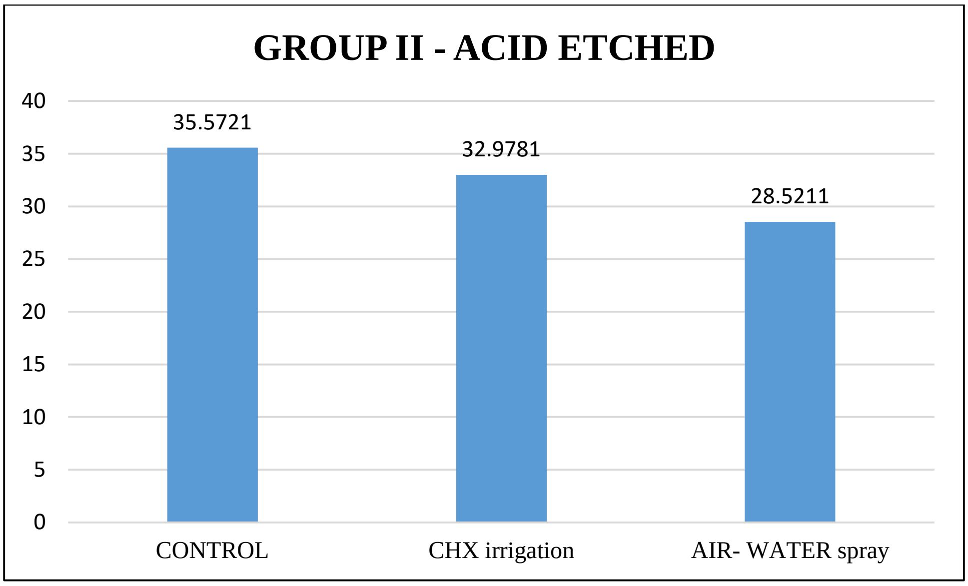


Graph 8: Mean area covered by plastic remnants after instrumentation and irrigation in Group I polished titanium discs

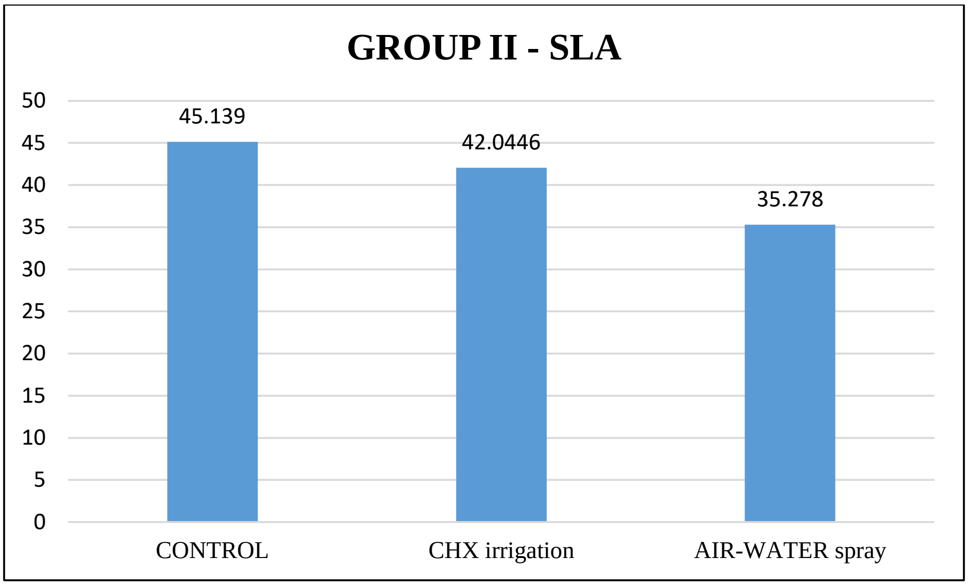
MEAN AREA OF PLASTIC REMNANTS AFTER INSTRUMENTATION AND IRRIGATION IN GROUP II



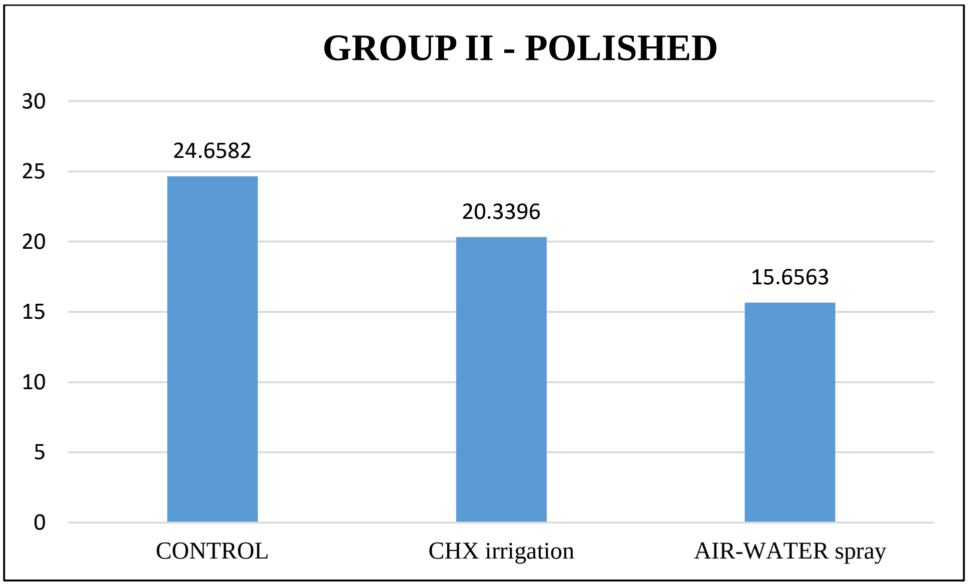
Graph 9: Mean area covered by plastic remnants after instrumentation and irrigation in Group II sand blasted titanium discs



Graph 10: Mean area covered by plastic remnants after instrumentation and irrigation in Group II acid etched titanium discs

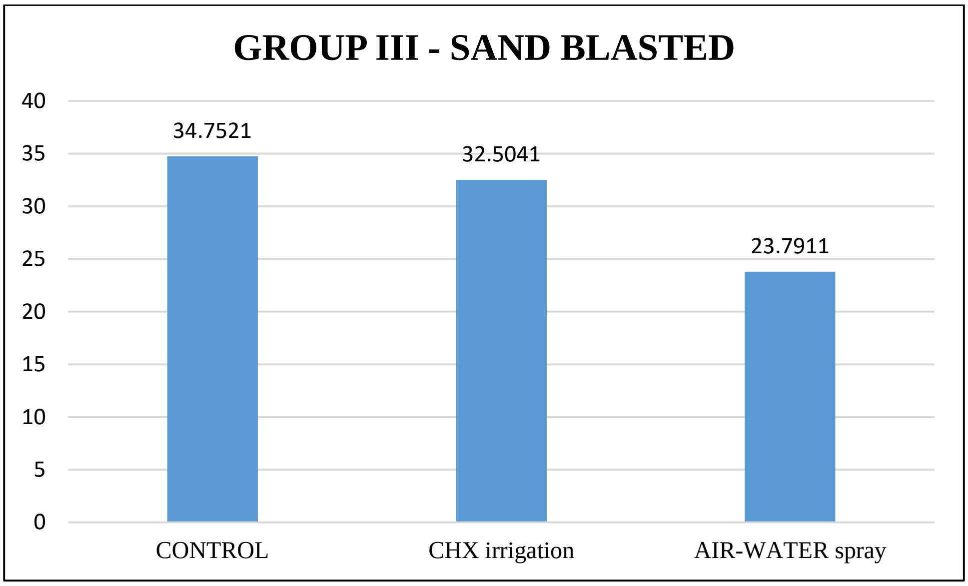


Graph 11: Mean area covered by plastic remnants after instrumentation and irrigation in Group II SLA titanium discs

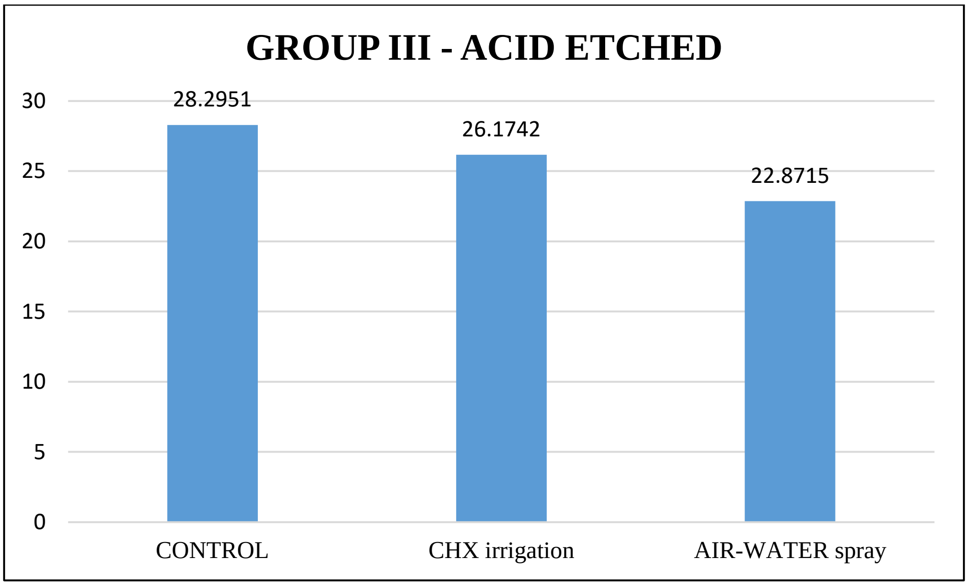


Graph 12: Mean area covered by plastic remnants after instrumentation and irrigation in Group II polished titanium discs

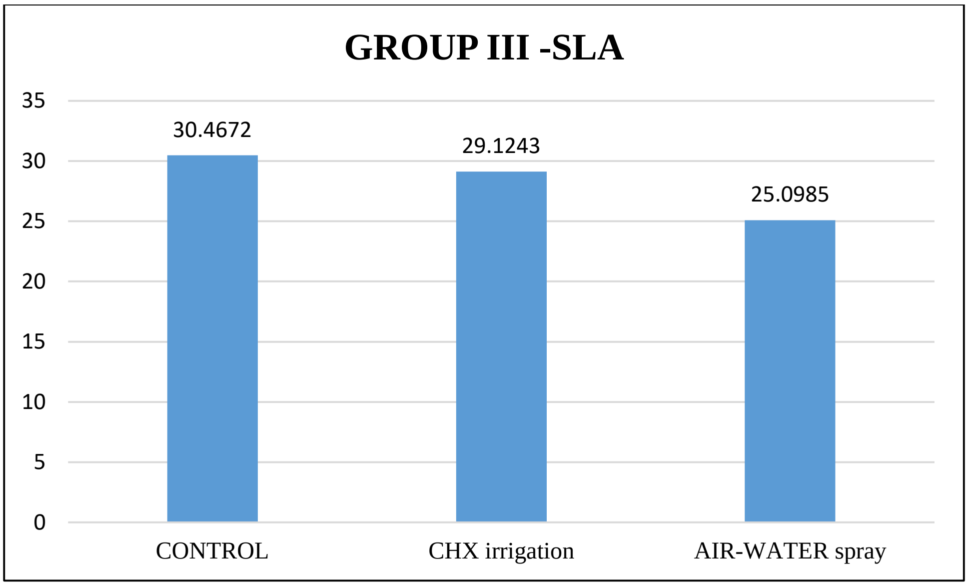
MEAN AREA OF PLASTIC REMNANTS AFTER INSTRUMENTATION AND IRRIGATION IN GROUP III



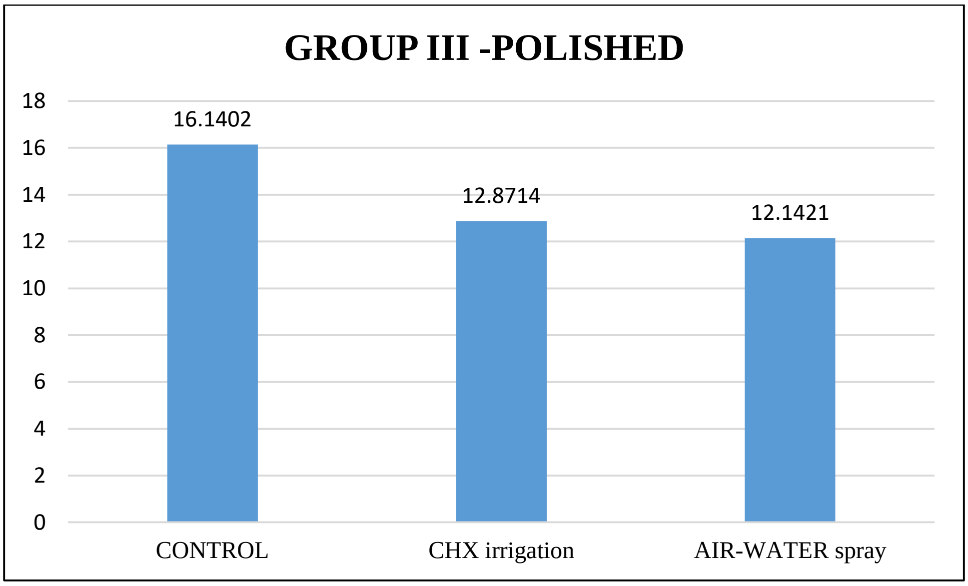
Graph 13: Mean area covered by plastic remnants after instrumentation and irrigation in Group III sand blasted titanium discs



Graph 14: Mean area covered by plastic remnants after instrumentation and irrigation in Group III acid etched titanium discs

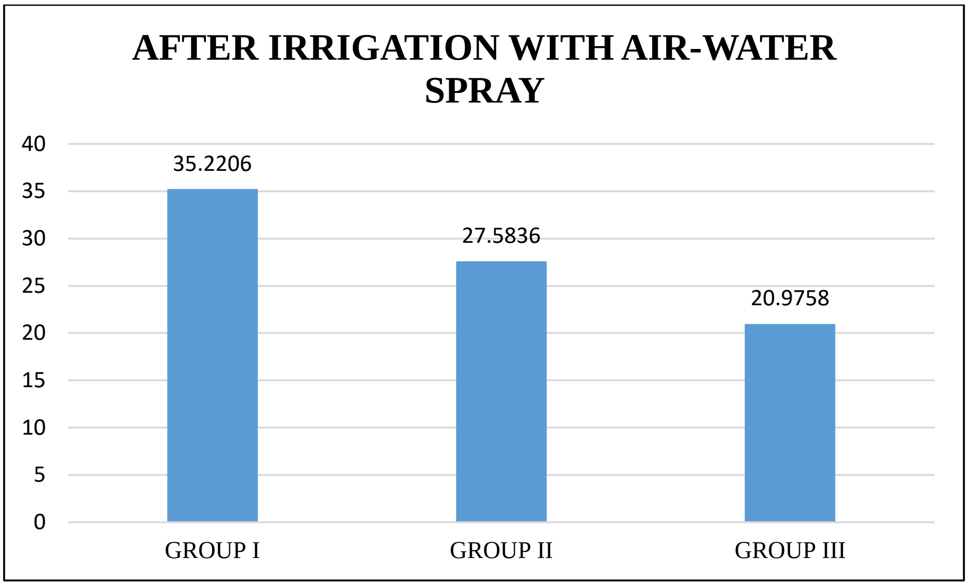


Graph 15: Mean area covered by plastic remnants after instrumentation and irrigation in Group III SLA titanium discs

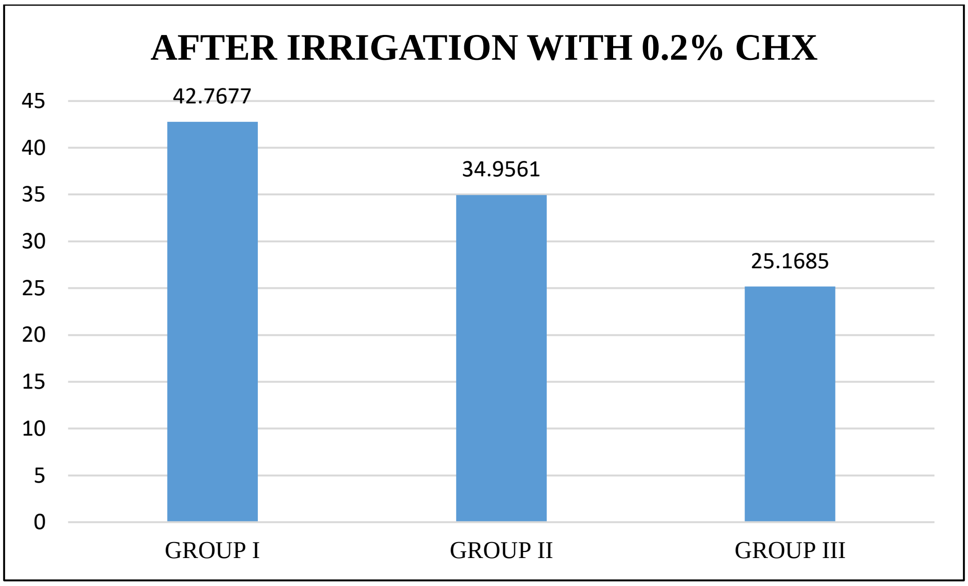


Graph 16: Mean area covered by plastic remnants after instrumentation and irrigation in Group III polished titanium discs

MEAN AREA COVERED BY PLASTIC REMNANTS IN 3 GROUPS AFTER IRRIGATION



Graph 17: Mean area covered by plastic remnants in 3 Groups after irrigation with air-water spray



Graph 18: Mean area covered by plastic remnants in 3 Groups after irrigation with 0.2% chlorhexidine

DISCUSSION

The treatment of peri-implant disease requires the complete debridement of micro-organisms from the implant surface, for which many techniques have been used and assessed. Current modalities include both non-surgical and surgical approaches. Non-surgical treatment approach includes mechanical cleansing, antiseptic therapy and antibiotic therapy of which surface debridement constitutes the basic element[1].

Various instruments have been used for surface debridement such as plastic instruments and titanium instruments, of which, several studies have advocated the use of plastic instruments considering the fact that it does not alter the surface roughness like stainless steel instruments[3,9]. However, it has been reported that instrumentation with plastic instruments, may leave behind plastic remnants[23] which may have far reaching consequences such as bacterial attachment, disruption in cell attachment and effectiveness of oral hygiene measures, thus, suggesting that the biocompatibility of the implant surfaces may be impaired by the plastic debris[5].

Various studies have demonstrated the presence of plastic remnants on the implant surface following instrumentation. Presence of plastic deposits were detected following instrumentation with plastic scaler on transmucosal abutments. These deposits appeared black on SEM and appeared to adhere to the irregular edges of the metal tags on the abutment surface[12]. Similarly, plastic debris were macroscopically visible following instrumentation with Teflon coated mechanical instruments and plastic curettes on rough surfaces such as titanium plasma sprayed implants[3,20]. These residual plastic debris were also found to have an effect on the growth of fibroblasts[18]. Instrumentation with Teflon coated ultrasonic and sonic scaler also left behind plastic remnants. It was observed that the plastic tip tended to melt at high power settings which stuck to the implant surface despite the use of a constant flow of water (30ml/min). This was seen under SEM as a bright flaring, which might be a build-up of electrical charge due to the non-conducting nature of the plastic deposit. It was also likely that the motion of the plastic tip against the edges of the screw thread of the dental implant may lead to more shredding of the plastic with greater deposits left behind[23]. Similarly, use of ultrasonic scaler with plastic PEEK tip on smooth machined surfaces and moderately roughened surfaces demonstrated particles embedded in the irregularities and crevices[34]. Presence of plastic debris was also found in the gap of the fixture abutment connection following instrumentation[35].

It has also been suggested that the biocompatibility of the implant surfaces may be impaired by the presence of these plastic debris. These debris may have an effect on the bacterial attachment. It is known that rough surfaces harbour 25 times more bacteria as compared to smooth surfaces[2]. Surface roughness promotes the colonisation because it shelters bacteria from clearance forces of salivary flow, chewing, swallowing and hygiene procedures, thereby allowing them to establish less reversible bindings[51]. The presence of plastic debris left behind after instrumentation might increase the surface roughness thereby resulting in increased plaque accumulation. It was found that titanium discs instrumented by plastic curette had increased residual plaque biofilm compared to control and discs treated with PEEK tip. This could be attributed by the fact that plastic debris were left behind by the plastic curette[52].

Plastic debris may also have an effect on the attachment of various cells such as epithelial cells, fibroblasts and osteoblasts. Effect of scaling procedures on epithelial cell growth on titanium surfaces was evaluated. Reduced epithelial growth on plastic scaled titanium discs was seen compared to control and stainless steel instrumented titanium discs. This was attributed by the authors to deposition of particles of plastic curette on the treated titanium discs[13]. It was found that fibroblasts grown on plastic curette instrumented titanium discs showed well spread polygonal morphology, typical of fibroblasts grown in favourable cell conditions[8]. Contradictory studies demonstrated reduced growth of fibroblasts on implants treated with plastic scaler when compared to control. Authors reported the presence of amorphous plastic like material between the cells[18]. The quality of the growth was also found to be impaired[54]. The effect on fibroblasts following instrumentation with carbon composite tip was also evaluated. Decreased proliferation of fibroblasts was seen on machined surfaces and increased proliferation was seen on rough (SLA) surface[53]. Surfaces treated with Vector ultrasonic scaler and PEEK tip was found to have reduced osteoblast cell attachment compared to Er:YAG treated implant surface and control. The authors attributed the reduced cell numbers to the cytotoxic effects of the fragments from the plastic tip. Surface modification also seemed to play a role in the cell attachment. It was found that the cell attachment was highest on SLA surfaces followed by titanium plasma sprayed and polished surfaces[52].

Thus, various surface modified implant surfaces does have a role in determining the amount of plastic remnants left behind after instrumentation with plastic instruments. With this background, the current study was conducted to quantify the amount of surface area covered by plastic remnants after instrumentation on various surface modified titanium discs and also to evaluate the efficacy of removal of these remnants after irrigation with 0.2% chlorhexidine and air-water spray.

In the current study, various surface modified titanium discs were instrumented with 1) Plastic curette- Hu Friedy 2) Carbon composite tip – Satelec and 3) PEEK tip (EMS) and the surface area covered by plastic remnants was assessed after instrumentation. It was found that the plastic curette left behind the maximum plastic debris which had a mean surface area of 47.38 ± 1.26% followed by the Carbon composite tip which had a mean surface area of 38.72 ± 1.03% and the least amount of plastic remnants were found with the PEEK tip with a surface area of 27.4 ± 7.49%. The difference between all the 3 groups were found to be statistically significant ($p<0.05$). A similar study was conducted by Yang et al[5] (2015) which demonstrated that the discs instrumented with plastic curette had the highest surface area of 17.7±3.2% covered by plastic remnants, which was in accordance with our study. Plastic curettes were followed by the PEEK tip which left behind plastic remnants with a surface area of 9.5±2.1% and the least by carbon composite tip, which occupied an area of 8.9±3.7%. The difference among the groups were found to be of statistical significance. ($p<0.05$).

The instrumented discs were then irrigated using air-water spray and 0.2% chlorhexidine. After irrigation, there was a decrease in surface area covered by plastic remnants in all discs and in all groups. After irrigation with air-water spray in plastic curette instrumented discs, the surface area covered by plastic remnants reduced from 47.38 ± 1.26% to 35.22 ± 8.65%. In carbon composite instrumented discs, the surface area covered by plastic remnants reduced from 38.72 ± 1.03% to 27.58 ± 7.94% and in the PEEK tip instrumented discs, the surface area covered by plastic remnants reduced from 27.41 ± 7.49% to 20.97 ± 5.71%. The difference among the 3 groups were found to be statistically significant. The results were in accordance with a study conducted by Yang et al[5] (2015) which demonstrated a reduction from 17.7 ± 3.2% to 11.4 ± 3.3% in plastic curette instrumented discs. In carbon composite tip instrumented discs, the reduction was from 8.9 ± 3.7% to 6.6 ± 2.4%. In discs instrumented by PEEK tip, the reduction was from 9.5 ± 2.1% to 8.0 ± 1.7%. The difference among the groups were of statistical significance.

The use of 0.2% chlorhexidine irrigation reduced the surface area covered by plastic remnants from 47.38 ± 1.26% to 42.76 ± 1.02% in the discs instrumented by plastic curette. In the carbon composite tip instrumented discs, the surface area covered by plastic remnants reduced from 38.72 ± 1.03% to 34.95 ± 1.01%. In the PEEK tip instrumented discs, the surface area covered by plastic remnants reduced from 27.41 ± 7.49% to 25.16 ± 8.03%. The difference among the 3 groups were found to be statistically significant ($p<0.05$). The results were in accordance with a study conducted by Yang et al[5] (2015) which demonstrated a reduction from 17.7 ± 3.2% to 15.5 ± 4.8% in plastic curette instrumented discs. In carbon composite tip instrumented discs, the reduction was from 8.9 ± 3.7% to 7.0 ± 3.8%. In discs instrumented by PEEK tip, the reduction was from 9.5 ± 2.1% to 7.9 ± 1.4%. The difference among the groups were of statistical significance ($p<0.05$).

No scientific literature yet has quantified and compared the plastic remnants on various surface modified titanium discs. It was found that sand blasted titanium discs had the most plastic remnants followed by SLA and acid etched. The least was found with polished titanium discs.

Overall, it was found that plastic remnants were present following instrumentation with all the plastic instruments and these remnants remained regardless of the irrigation method used. Out of which, air-water spray had a better irrigating effect in removing the plastic remnants after instrumentation when compared to 0.2% chlorhexidine irrigation. This study highlighted the difficulty of removing all remnants with air-water spray and 0.2% chlorhexidine. This study also suggests that confocal microscopy can be a useful and reproducible method for quantification of plastic remnants on various modified implant surfaces. Within the limitations of this study, we need to emphasize the importance of the irrigants used and the method of irrigation to ensure the complete removal of plastic remnants after instrumentation which might affect the biocompatibility of the implant surface. It is suggested that, air-water spray may be an efficient method of irrigation to remove the plastic remnants and could be used as one of the methods of debridement of implant surface.

SUMMARY & CONCLUSION

Various procedures and instruments have been proposed to reduce the number of pathogenic species and consequently improve or preserve periodontal health around titanium implants. The procedures usually involves the removal of microbial deposits and debris without altering the surface of the implant in such a way as to adversely affect the biocompatibility. Plastic instruments have been used as it does not alter the surface. But instrumentation with plastic instruments have been known to leave behind some plastic remnants. These remnants can impair the biocompatibility of the implant surface.

Hence, this study was conducted to quantify the surface area covered by plastic instruments after instrumentation using different plastic instruments on various surface modified titanium discs. The surface area covered by plastic debris was quantified using a confocal laser scanning microscope and MATLAB software.

Results obtained in this study are:

i. Overall, Group I (Plastic curette-Hu Friedy) had the maximum surface area covered by plastic remnants followed by Group II (Carbon composite tip-Satelec) and the least was found with Group III (PEEK tip-EMS) after instrumentation and irrigation. The difference between the groups was found to be statistically significant.

ii. Among the various surface modifications, it was found that sand blasted titanium discs retained the maximum plastic remnants followed by SLA. Polished titanium discs retained the least amount of plastic remnants as compared to sand blasted and SLA surfaces. The difference among the groups was found to be statistically significant.

iii. Following irrigation with air-water spray and 0.2% chlorhexidine, it was found that Group I retained more plastic remnants compared to Group II and Group III.

iv. Overall, it was found that plastic remnants remained after instrumentation, regardless of the irrigation method used. But it was found that the air-water spray had a better irrigating effect in removing the plastic remnants after instrumentation when compared to 0.2% chlorhexidine irrigation.

From the results obtained in the present study, we can conclude that plastic remnants remained after instrumentation, irrespective of the instrument used. Among all the instruments used, PEEK tip (EMS) left behind the least plastic remnants after instrumentation and irrigation. Even though, air-water spray had a better irrigating effect in removing the remnants, complete removal was not possible. Further research is warranted to evaluate the irrigants and method of irrigation to ensure complete removal of plastic remnants which might impair the biocompatibility of the implant surface.

BIBLIOGRAPHY

1. Renvert S, Roos-Jansaker AM, Claffey N. Non-surgical treatment of peri-implant mucositis and peri-implantitis: a literature review. *J Clin Periodontol* 2008; 35:305–15.
2. Quirynen M, Van Der Mei HC, Bollen CML, Schotte A, Marechal M, Doornbusch GI. An in vivo study of the influence of the surface roughness of implants in microbiology of supra- and subgingival plaque. *J Dent Res* 1993;72:1304–9.
3. Ruhling A, Kocher T, Kreusch J, Plagmann HC. Treatment of subgingival implant surfaces with Teflon-coated sonic and ultrasonic scaler tips and various implant curettes. An in vitro study. *Clin Oral Implants Res* 1994;5: 19–29.
4. Schwarz F, Rothamel D, Sculean A, Georg T, Scherbaum W and Becker J. Effects of an Er:YAG laser and the vector ultrasonic system on the biocompatibility of titanium implants in cultures of human osteoblast-like cells. *Clin Oral Implants Res* 2003;14: 784–92.
5. Yang SM, Park JB, Ko Y. Use of confocal microscopy for quantification of plastic remnants on rough titanium after instrumentation and evaluation of efficacy of removal. *Int J Oral Maxillofac Implants* 2015; 30: 519–25.
6. Piruska A, Nikcevic I, Lee SH. The autofluorescence of plastic materials and chips measured under laser. *Irradiation.Lab chip* 2005; 5:1348-54.
7. Fox SC, Moriarty JD, Kusy RP. The effects of scaling a titanium implant surface with metal and plastic instruments: an in vitro study. *J Periodontol* 1990; 61: 485–90.
8. Dmytryk JJ, Fox SC, Moriarty JD. The effect of scaling titanium surfaces with metal and plastic instruments on cell attachment. *J Periodontol* 1990; 61:491–96.
9. Speelman JA, Collaert B, Klinge B. Evaluation of different methods to clean titanium abutments. A scanning electron microscopic study. *Clin Oral Implants Res* 1992;3: 120– 27.

10. Homiak AW, Cook PA, De Bore J. Effects of hygiene on titanium abutments: A scanning electron microscopy study. *J Prosthet Dent* 1992;67:364-69.
11. McCollum J, O'Neal RB, Brennan WA, Van Dyke TE, Horner JA. The effect of titanium implant abutment surface irregularities on plaque accumulation in vivo. *J Periodontol* 1992; 63(10):802-5.
12. Brookshire FV, Nagy WW, Dhuru VB, Ziebert GJ, Chada S. The qualitative effects of various types of hygiene instrumentation on commercially pure titanium and titanium alloy implant abutments: an in vitro and scanning electron microscope study. *J Prosthet Dent* 1997; 78(3):286-94.
13. Kuempel DR, Johnson GK, Zaharias RS, Keller JC. The effects of scaling procedures on epithelial cell growth on titanium surfaces. *J Periodontol* 1995;66(3):228-34.
14. Meschenmoser A, D'Hoedt B, Meyle J, Elssner G, Korn D, Hammerle H, Schulte W. Effects of various hygiene procedures on the surface characteristics of titanium abutments. *J Periodontol* 1996; 67: 229–35.
15. Matarasso S, Quaremba G, Coraggio F, Vaia E, Cafiero C, Lang NP. Maintenance of implants: an in vitro study of titanium implant surface modifications subsequent to the application of different prophylaxis procedures. *Clin Oral Implants Res* 1996; 6: 64–72.
16. Hallmon WW, Waldrop TC, Meffert RM, Wade BW. A comparative study of the effects of metallic, non-metallic and sonic instrumentation on titanium abutment surfaces. *Int J Oral Maxillofac Implants* 1996;11:96-100.
17. Mengel R, Buns CE, Mengel C, Flores-de-Jacoby L. An in vitro study of treatment of implant surface with different instruments. *Int J Oral Maxillofac Implants* 1998; 13:91-96.

18. Augthun M, Tinschert J, Huber A. In vitro studies on the effect of cleaning methods on different implant surfaces. *J Periodontol* 1998;69: 857-64.

19. Sato S, Kishida M, Ito K. The comparative effect of ultrasonic scalers on titanium surfaces: An in vitro study. *J Periodontol* 2004; 75:1269-73.

20. Karring ES, Stavropoulos A, Ellegaard B, Karring T. Treatment of peri-implantitis by the Vectors system. A pilot study. *Clin. Oral Imp Res* 2005; 16: 288–93.

21. Ramaglia, L, Di Lauro A.E, Morgese F, Squillace A. Profilometric and standard error of the mean analysis of rough implant surfaces treated with different instrumentations. *Implant Dent* 2006; 15: 77–82.

22. Kawashima H, Sato S, Kishida M, Yagi H, Matsumoto K, Ito K. Treatment of titanium dental implants with three piezoelectric ultrasonic scalers: an in vivo study. *J Periodontol* 2007; 78: 1689–94.

23. Mann M, Parmar D, Walmsley AD, Lea SC. Effect of plastic-covered ultrasonic scalers on titanium implant surfaces. *Clin Oral Implants Res* 2012; 23: 76–82.

24. Baek SH, Shon WJ, Bae KS, Kum KY, Lee WC, Park YS. Evaluation of the safety and efficiency of novel metallic ultrasonic scaler tip on titanium surfaces. *Clin Oral Implants Res* 2011; 1–6

25. Sahm N, Becker J, Santel T, Schwarz F. Non-surgical treatment of peri-implantitis using an air-abrasive device or mechanical debridement and local application of chlorhexidine: a prospective, randomized, controlled clinical study. *J Clin Periodontol.* 2011;38(9):872-8.

26. Schmage P, Thielemann J, Nergiz I, Scorziello TM, Pfeiffer P. Effects of 10 cleaning instruments on four different implant surfaces *Int J Oral Maxillofac Implants* 2011;27: 308–17.

27. Park JB, Kim N, Ko Y. Effects of ultrasonic scaler tips and toothbrush on titanium disc surfaces evaluated with confocal microscopy. *J Craniofac Surg* 2012 ;23(5):1552-8.

28. Fakhravar B, Khocht A, Jefferies SR, Suzuki JB. Probing and scaling instrumentation on implant abutment surfaces: an in vitro study. *Implant dent* 2012;21(4):311-6.

29. Unursaikhan O, Lee JS, Cha JK, Park JC, Jung UW, Kim CS, Cho KS, Choi SH. Comparative evaluation of roughness of titanium surfaces treated by different hygiene instruments. *J Periodontal Implant Sci* 2012;42:88–94.

30. Park JB, Jang YJ, Koh M, Choi BK, Kim KK, Ko Y. In vitro analysis of the efficacy of ultrasonic scalers and a toothbrush for removing bacteria from resorbable blast material titanium disks. *J Periodontol* 2013;84: 1191–98.

31. Park JB, Jang YJ, Choi BK, Kim KK, Ko Y. Treatment with various ultrasonic scaler tips affects efficiency of brushing of SLA titanium discs. *J Craniofac Surg* 2013;24(2):119-23.

32. Blasi A, Iorio-Siciliano V, Pacenza C, Pomingi F, Matarasso S, Rasperini G. Biofilm removal from implants supported restoration using different instruments: a 6-month comparative multicenter clinical study. *Clin Oral Impl Res* 2014;27(2):1–6.

33. Schmage P, Kahili F, Nergiz I, Scorziello TM, Platzer U, Pfeiffer P. Cleaning effectiveness of implant prophylaxis instruments. *Int J Oral Maxillofac Implants* 2014;29(2).

34. Tawse-Smith A, Atieh MA, Tompkins G, Duncan WJ, Reid MR, Stirling CH. The effect of piezoelectric ultrasonic instrumentation on titanium discs: a microscopy and trace elemental analysis in vitro study. *Int J Dent Hyg* 2016;14(3):191-201.

35. Bertoldi C, Lusuardi D, Battarra F, Sassatelli P, Spinato S, Zaffe D. The maintenance of inserted titanium implants: invitro evaluation of exposed surfaces cleaned with three different instruments. *Clin Oral Impl Res* 2017;28(1):57-63.

36. Ronay V, Merlini A, Attin T, Schmidlin PR, Sahrmann P. In vitro cleaning potential of three implant debridement methods. Simulation of the non-surgical approach. *Clin Oral Impl. Res* 2017;28(2):151-5.
37. Al-Hashedi AA, Laurenti M, Benhamou V, Tamimi F. Decontamination of titanium implants using physical methods. *Clin Oral Impl Res* 2017;28(8):1013-21.
38. Schmidt KE, Auschill TM, Heumann C, Frankenberger R, Eick S, Sculean A, Arweiler NB. Influence of different instrumentation modalities on the surface characteristics and biofilm formation on dental implant neck, in vitro. *Clin Oral Impl Res* 2017;28(4):483-90.
39. Zablotsky MH, Diedrich DL, Meffert RM. Detoxification of endotoxin- contaminated titanium and hydroxyapatite-coated surfaces utilizing various chemotherapeutic and mechanical modalities. *Implant Dent* 1992;1(2):154-8.
40. Dennison DK, Huerzeler MB, Quinones C, Caffese RG. Contaminated implant surfaces: an *in vitro* comparison of implant surface coating and treatment modalities for decontamination. *J Periodontol* 1994; 65(10): 942-8.
41. Felo A, Shibly O, Ciancio SG, Lauciello FR, Ho A. Effects of subgingival chlorhexidine irrigation on peri-implant maintenance. *Am J Dent* 1997; 10: 107-10.
42. Porras R, Anderson GB, Caffesse R, Narendran S, Trejo PM. Clinical response to 2 different therapeutic regimens to treat peri-implant mucositis. *J Periodontol* 2002; 73(10):1118-25.
43. Trejo PM, Bonaventura G, Weng D, Caffesse RG, Bragger U, Lang NP. Effect of mechanical and antiseptic therapy on peri-implant mucositis: an experimental study in monkeys. *Clin Oral Implants Res* 2006; 17(3): 294-304.

44. Sennhenn-Kirchner S, Wolff N, Klaue S, Mergeryan H, Borg-von MZ. Decontamination efficacy of antiseptic agents on in vivo grown biofilms on rough titanium surfaces. *Quintes Int* (Berlin, Germany: 1985) 2009;40(10):80-8.
45. Gosau M, Hahnel S, Schwarz F, Gerlach T, Reichert TE, Burgers R. Effect of six different peri-implantitis disinfection methods on in vivo human oral biofilm. *Clin Oral Impl Res* 2010;21: 866–72.
46. Thone-Muhling M, Swierkot K, Nonnenmacher C, Mutters R, Flores-de-Jacoby L, Mengel R. Comparison of two full-mouth approaches in the treatment of peri-implant mucositis: a pilot study. *Clin Oral Impl Res* 2010;21: 504–12.
47. Ntrouka VI, Slot DE, Louropoulou A, Van der Weijden F. The effect of chemotherapeutic agents on contaminated titanium surfaces: a systematic review. *Clin. Oral Impl Res* 2011; 22(7):681-90.
48. Bürgers R, Witecy C, Hahnel S, Gosau M. The effect of various topical peri-implantitis antiseptics on Staphylococcus epidermidis, Candida albicans, and Streptococcus sanguinis. *Arch Oral Biol* 2012; 57(7): 940-7.
49. Charalampakis G, Ramberg P, Dahlen G, Berglundh T, Abrahamsson I. Effect of cleansing of biofilm formed on titanium discs. *Clin Oral Impl Res* 2015;26(8):931-6.
50. Lee BS, Shih KS, Lai CH, Takeuchi Y, Chen YW. Surface property alterations and osteoblast attachment to contaminated titanium surfaces after different surface treatments: An in vitro study. *Clin Implant Dent Relat Res* 2018;00:1–9.
51. Rimondini L, Farè S, Brambilla E, Felloni A, Consonni C, Brossa F, Carrassi A. The effect of surface roughness on early in vivo plaque colonization on titanium. *J Periodontol* 1997;68(6):556-62.
52. Schwarz F, Sculean A, Romanos G, Herten M, Horn N, Scherbaum W, Becker J. Influence of different treatment approaches on the removal of early plaque biofilms and

the viability of SAOS2 osteoblasts grown on titanium implants. *Clin Oral Investig* 2005;9(2):111-7.

53. Cao J, Wang T, Pu Y, Tang Z, Meng H. Influence on proliferation and adhesion of human gingival fibroblasts from different titanium surface decontamination treatments: An in vitro study. *Arch oral biol* 2018;87:204-10.

54. Kim B, Park, Ko Y. Growth of human gingival fibroblasts on plastic debris remaining on rough surface titanium after instrumentation. *Clin Oral Impl Res* 2016;27 (13) 75.